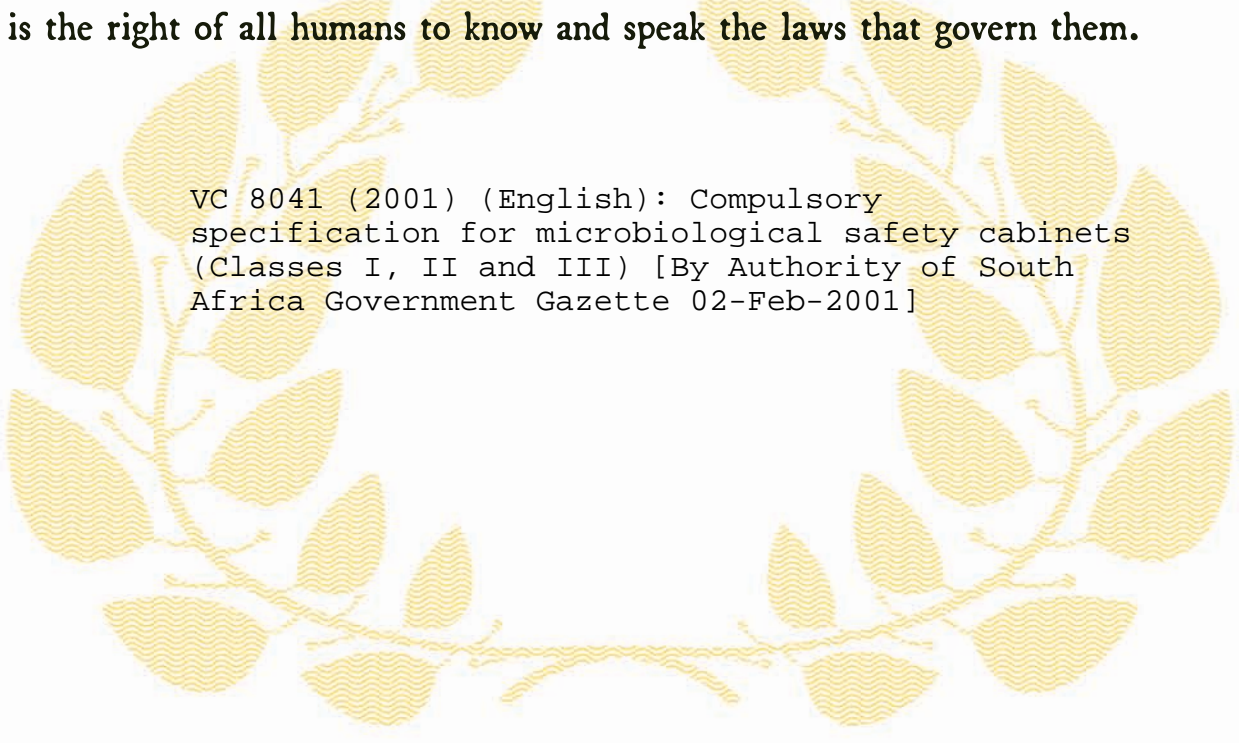




# *Republic of South Africa*

## EDICT OF GOVERNMENT

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VC 8041 (2001) (English): Compulsory  
specification for microbiological safety cabinets  
(Classes I, II and III) [By Authority of South  
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Compulsory Specification for

**Microbiological safety cabinets (Classes I, II and III)**

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## GOVERNMENT NOTICES GOEWERMENTSKENNISGEWINGS

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### DEPARTMENT OF TRADE AND INDUSTRY DEPARTEMENT VAN HANDEL EN NYWERHEID

No. R. 93

2 February 2001

STANDARDS ACT, 1993

#### THE COMPULSORY SPECIFICATION FOR MICROBIOLOGICAL SAFETY CABINETS (CLASSES I, II AND III)

I, Alexander Erwin, Minister of Trade and Industry, hereby under section 22 (1) (a) (i) of the Standards Act, 1993 (Act No. 29 of 1993), and on the recommendation of the Council of the South African Bureau of Standards, amend the compulsory specification for microbiological safety cabinets (classes I, II and III), as set out in the Schedule, with effect from the date of two months after the date of publication of this notice.

**A. ERWIN****Minister of Trade and Industry**

## SCHEDULE

### COMPULSORY SPECIFICATION FOR MICROBIOLOGICAL SAFETY CABINETS (CLASSES I, II AND III)

#### 1 Scope

**1.1** This specification covers requirements for the construction, fittings and pre-installation and post-installation performance of class I, class II and class III microbiological safety cabinets (also known as biological safety cabinets) intended to protect the operator and the environment from hazardous microbiological materials and (if so required by the customer or user or both), organic toxins and non-corrosive volatile organic agents.

NOTE – Microbiological safety cabinets are not intended to provide protection against corrosive chemical or radioactive materials.

**1.2** The specification does not cover the actual design of a safety cabinet and in no way restricts new design, provided that a microbiological safety cabinet of a new design complies with the requirements for materials, reliability, performance and safety given in this specification.

NOTE – Microbiological safety cabinets of class I, class II and class III should not be confused with laminar flow clean workstations that usually discharge horizontally and vertically towards the operator and that do not provide protection for an operator, but can even increase exposure to airborne hazards.

#### 2 Definitions and abbreviations

For the purposes of this specification, the following definitions and abbreviations apply:

##### 2.1 Definitions

**2.1.1 accessible:** Able to be exposed for proper and thorough cleaning and visual inspection, with the use of simple tools such as a screwdriver, pliers or a spanner.

**2.1.2 barrier air; face air:** Atmospheric air sucked from the room environment through the work-access aperture of the cabinet, to create an air barrier across the aperture, through which particles cannot escape from the cabinet to the outside atmosphere.

**2.1.3 cabinet:** A microbiological safety cabinet of class I, class II or class III, as applicable.

**2.1.4 cleanable:** Accessible and of such material and finish and so manufactured that soil can be removed effectively by normal cleaning methods.

**2.1.5 construction:** The manufacture, assembly of subunits (where applicable) and installation of the microbiological safety cabinet.

**2.1.6 disinfection; decontamination:** The removal or inactivation of infectious agents or the removal or neutralizing of toxic agents.

**2.1.7 glare:** A condition of vision in which there is discomfort, or a reduction in the ability to distinguish significant objects, or both, due to an unsuitable distribution or range of luminances or to extreme contrasts, simultaneous or successive, in the visual field.

**2.1.8 hazard; biohazard; hazardous materials:** Infectious agents that present a real or potential risk to the well-being of human beings, animals or plants, either directly through infection or indirectly through contamination of the environment.

**2.1.9 HEPA-filter:** A high-efficiency particulate air filter.

**2.1.10 internal work surface:** The interior of the work space that can easily be wiped.

**2.1.11 readily accessible; easily accessible:** Easily exposed for proper and thorough cleaning and for visual inspection without the use of any tool.

**2.1.12 readily removable:** Able to be (capable of being) taken away from the main unit without the use of any tool.

**2.1.13 removable:** Able to be (capable of being) taken away from the main unit with the use of simple tools such as a screwdriver, pliers or a spanner.

**2.1.14 resistant:** Descriptive of materials that maintain their original surface characteristics under conditions other than those intended for normal use.

**2.1.15 sealed:** Having no openings that will allow the entry or leakage of water or gas.

**2.1.16 smooth:** Having a surface free from pits and inclusions.

**2.1.17 toxic:** Descriptive of agents that have an adverse physiological effect on biological systems.

**2.1.18 toxins:** Agents that have an adverse physiological effect on biological systems.

**2.1.19 work space:** That part of the interior of the cabinet, within which handling of the hazardous material can safely be carried out.

## 2.2 Abbreviations

**2.2.1 AISI:** American Iron and Steel Institute.

**2.2.2 DOP:** Di-octylphthalate.

## 3 General requirements

### 3.1 Class

A cabinet shall be of one of the following classes:

- a) **class I:** A partially enclosed cabinet that is so constructed that air flows inwards through the work-access aperture and away from the operator. The exhaust air is filtered through a HEPA-filter before being discharged from the cabinet.

**WARNING — These cabinets shall not be used as, or confused with, fume cabinets which are intended for chemical procedures.**

**NOTE —** Class I cabinets provide protection for personnel and the environment against ordinary or potentially hazardous microbiological agents, i.e. at risk levels associated with agents that cause disease in human beings, animals or plants, provided that the usual precautions in handling microbiological material are observed. The level of competence required of personnel who handle material in these cabinets should be that of personnel formally trained as microbiologists.

- b) **class II:** A partially enclosed cabinet that is so constructed that the air in the work space is flushed with a clean, HEPA-filtered, unidirectional downward flow of air, and the escape of particles from the work space is prevented by an inward flow of air through the work-access aperture. The exhaust air is filtered through a HEPA-filter before being discharged from the cabinet.

NOTE – Class II cabinets provide protection for personnel, the environment and the product against ordinary or potentially hazardous microbiological agents, i.e. at risk levels associated with agents that cause disease in human beings, animals or plants, provided that the usual precautions in handling microbiological materials are observed. The level of competence required of personnel who handle material in these cabinets should be that of personnel formally trained as microbiologists.

- c) **class III:** A totally enclosed, ventilated cabinet of gastight construction that is so constructed that the operator is separated from the work space by a physical barrier, and the work space is so flushed with HEPA-filtered air under negative pressure that the escape of particles from the work space is highly unlikely. The exhaust air is filtered through a HEPA-filter before being discharged from the cabinet to the outside atmosphere through an airtight duct.

NOTE – Class III cabinets provide protection for personnel and the environment against special and extremely hazardous microbiological agents, i.e. at risk levels associated with agents that are highly infectious or toxic to human beings, animals and plants, and that can cause dangerous disease, or at risk levels associated with agents that cause genetic mutations or that can have a synergistic effect with other materials. The cabinet also controls airborne contamination that might be detrimental to the work in the work space. The level of competence required of personnel who handle material in these cabinets should be that expected of personnel formally trained as microbiologists and who have also received proper training in the handling of extremely dangerous agents.

## 3.2 Dimensions

### 3.2.1 External dimensions

The overall dimensions of a cabinet, excluding the readily removable parts, shall be such that it can pass through a standard single doorway of nominal height and width 2,0 m and 0,78 m respectively.

### 3.2.2 Work space dimensions

In the case of class I and class II cabinets, the width of the work space shall not exceed 1 900 mm and the depth shall be in the range 500 mm to 700 mm. The height of the work space shall be at least 550 mm. The volume of the work space shall be not less than 0,2 m<sup>3</sup> and not more than 0,75 m<sup>3</sup>.

## 3.3 Outer shell (main structure) – Material and construction

### 3.3.1 General

3.3.1.1 A cabinet shall be constructed of glass or metal that is deemed to be corrosion resistant when tested in accordance with 6.11. If stainless steel is used, it shall be of at least AISI Grade 304 and the requirement for corrosion resistance shall not apply.

3.3.1.2 There shall be no cracks and surface defects, including ineffective mating with gasket surfaces or other sealing devices. All structural joints that are not welded shall be sealed with a suitable sealant that is not liable to crack. The gasket or sealing material shall not be used as a structural material for any joint or connection of the cabinet or any of its panels.

### 3.3.2 Stability of the cabinet

When a free-standing cabinet is tested in accordance with 6.14, the rear bottom edge of the cabinet shall not lift off the surface on which it rests by more than 1,6 mm when a torque of 700 Nm is applied at the centre of the rear top edge. In the case of a cabinet installation that does not provide this degree of stability, provision shall be made for stabilizing the cabinet by appropriate means to ensure compliance.



### 3.3.3 Windows

Windows shall be of safety glazing material that complies with the performance requirements of SABS 1263-1:1986, *Safety and security glazing materials for buildings – Part 1: Safety performance of glazing materials under human impact*, as published by Government Notice No. 1851 of 1 December 1995 or of other suitable materials that are resistant to ultraviolet rays and that perform at least as well as the safety glazing material.

### 3.3.4 Access panels

Removable access panels or covers shall be provided for the maintenance or removal (or both) of filters, blowers, motors, lighting, electrical components and plumbing. When access panels or covers are in place, their seals shall prevent leakage of contaminated air to the surrounding atmosphere. Physical means to position and support large access panels or covers shall be provided to facilitate safe fitting and removal. Fastenings shall not compromise the integrity of the outer shell of cabinets. Fastenings of class III cabinets shall not penetrate the inner or outer shell. There shall be effective mating of the access panels or covers with the gasket surfaces.

NOTE – Access panels to contaminated zones should be removed only after the whole cabinet has been decontaminated.

### 3.3.5 Tracks and guides

All tracks and guides for doors, windows, access panels and covers shall be so constructed and installed as to minimize the collection of foreign matter and to facilitate cleaning.

### 3.3.6 Access points to the exhaust duct (class III cabinets)

Class III cabinets shall have access points to the exhaust duct to facilitate the measurement of airflow.

## 3.4 Work space — Material and construction

### 3.4.1 General

**3.4.1.1** The work space, excluding the viewing window, but including the work floor and its associated structures, the sump and grills, where applicable, shall be constructed entirely of a suitable metal that, when tested in accordance with 6.11, is deemed to be corrosion resistant. If stainless steel is used, it shall be of at least AISI Grade 304 and the requirement for corrosion resistance shall not apply.

**3.4.1.2** When an internal work surface (see 2.1.10) is tested in accordance with 6.13 and the surface is compared with untreated areas, there shall be no visible effect other than a slight change of gloss or discoloration.

NOTE – The resistance of the surface to special chemical solutions which are intended to be used, should also be evaluated.

**3.4.1.3** The surfaces shall be smoothly finished and cleanable (see 2.1.4), and shall be such that glare (see 2.1.7) from lighting (see 3.4.6.2) is avoided.

**3.4.1.4** In order to prevent penetration by microorganisms, all welds shall be ground flush and dressed. Joints, cracks and crevices in the work space shall be effectively sealed with a suitable sealant which is resistant to solvents and to normal disinfectants.

**3.4.1.5** There shall be no sharp projections within the work space.

### 3.4.2 Internal corners and angles

All internal corners and angles in the work space shall be

- a) free from cracks and crevices, and
- b) designed to facilitate cleaning and disinfection.

### **3.4.3 Viewing window**

**3.4.3.1** The viewing window shall consist of a transparent panel that complies with the requirements of 3.3.3 and that can be opened, in class I and class II cabinets only, to allow access to the work space.

**3.4.3.2** The viewing window shall form the front boundary of the clean air environment and shall not disrupt the laminar pattern of airflow.

**3.4.3.3** Apart from being optically clear and not adversely affected by accepted cleaning methods, the size, position and angle of the viewing window shall allow a clear and unobstructed view into the work space when the operator is seated centrally in front of the cabinet.

**3.4.3.4** When a class II cabinet is tested in accordance with 6.5, all seals around the top and sides of the viewing window shall have a DOP aerosol penetration not exceeding 0,03 %.

### **3.4.4 Work-access aperture (class I and class II cabinets only)**

The edges of the work-access aperture shall be so formed as to minimize air turbulence at the entry. The vertical dimension of the aperture shall be in the range 200 mm to 250 mm.

### **3.4.5 Work-access aperture cover (class I and class II cabinets only)**

A cover to fit the work-access aperture shall be provided in order to seal the cabinet during decontamination.

### **3.4.6 Work space illumination**

**3.4.6.1** The work space shall be illuminated by fluorescent lamps. The lamps and accessories shall be outside the work space. Replacement and maintenance of the lamps and accessories shall be carried out from the outside of the cabinet without compromising the integrity of the work space.

**3.4.6.2** The lamps shall be so positioned that their reflections do not impede the visibility through the window. The operator's eyes shall be shielded from direct radiation.

**3.4.6.3** When determined in accordance with 6.2, the average illuminance at the work surface shall be at least 800 lx (800 lumens per square metre). No single illuminance reading shall differ from the average illuminance by more than 20 %.

**3.4.6.4** Ultraviolet lamps shall not be installed as integral parts of the cabinet.

### **3.4.7 Control gear**

All control gear shall be accessible from the outside of the cabinet without the integrity of the plenums or biohazard safety barriers being affected. Control gear shall be so mounted and so sealed that there is no air leakage into the atmosphere, and that the control gear cannot become contaminated by contaminated air.

### **3.4.8 Screens**

In class I and class II cabinets, one or more screens shall be provided on the return air manifold to prevent any loose material from being drawn from the work space into the motor blower or the HEPA-filter housings. The screen(s) shall register in position without the need for fastening. The finish of the screen(s) shall be smooth to facilitate cleaning and disinfection.

### **3.4.9 Gas fittings**

**3.4.9.1** If the work space of class I and class II cabinets has a supply of flammable gas (for example, for bunsen burners) this supply shall be controlled by means of a solenoid valve that will allow the flow of gas only when the motor blowers are switched on.

**3.4.9.2** In order to reduce explosion hazard, the solenoid valve shall be such that it has to be manually reset after any interruption of the power supply.

**3.4.9.3** Class III cabinets shall not have gas fittings.

## **3.5 Air filters**

### **3.5.1 Recirculating, inlet and exhaust filters**

#### **3.5.1.1 Filter types**

The installed HEPA-filters shall have a volumetric rate (airflow rate), specified by the manufacturer of the filter, at least equal to the maximum rate necessary for the applicable part of the cabinet. Each HEPA-filter shall have been individually tested for filtration efficiency and filter integrity in accordance with an internationally acceptable standard at the manufacturer's designed volumetric rate.

#### **3.5.1.2 Filter frame**

All HEPA-filters shall have frames manufactured from metal that is corrosion resistant or protected against corrosion. Fluid or grease seals shall not be used.

#### **3.5.1.3 Filter installation**

**3.5.1.3.1** When the installed, recirculating, supply and exhaust HEPA-filters are tested in accordance with 6.4, there shall be no aerosol penetration exceeding 0,03 %.

**3.5.1.3.2** Gauges or manometers shall be fitted to monitor the pressure drop across the HEPA-filters. At least one gauge shall be provided.

**3.5.1.3.3** The cabinet's filter aperture(s) shall match the filter frame exactly.

**3.5.1.3.4** The filters shall not be fixed in place by means of adhesives or agents that solidify.

**3.5.1.3.5** Petroleum jelly shall not be used on the seals.

**3.5.1.3.6** The HEPA-filter gasket material shall be cellular, sheet or moulded rubber, or closed-cell expanded neoprene.

#### **3.5.1.4 Access to filters and sampling ports**

**3.5.1.4.1** Access shall be provided to facilitate servicing and determination of the integrity of filters and seals.

**3.5.1.4.2** Where necessary, sampling ports for determining the 100 % datum concentration of the DOP challenge aerosol shall be provided for each HEPA-filter positive-pressure plenum, and the ports shall be connected by a tube of internal diameter at least 15 mm, to accessible positions in the negative-pressure plenum.

**3.5.1.4.3** Each sampling port shall have a suitable, readily removable closure. Tubes and closures shall not penetrate the outer shell of the cabinet.

#### **3.5.1.5 Filter sealing plates**

Sealing plates for both the inlet and the exhaust (where applicable) opening shall be provided to facilitate fumigation and disinfection. Where these plates are provided to seal the filters, they shall be fitted externally over the filters and shall provide an effective seal, to ensure that the filters are also decontaminated during fumigation.

### **3.5.1.6 Protection**

A removable perforated guard shall be provided in the exhaust opening to protect the HEPA-filter from mechanical damage and shall be so arranged that the discharge of air is not obstructed.

### **3.5.2 Prefilters**

**3.5.2.1** In order to extend the life of the HEPA-filters, a prefilter that is readily accessible shall be fitted upstream of the HEPA-filters.

**3.5.2.2** When determined in accordance with 6.12, this prefilter shall have an average arrestance of 80 %.

**3.5.2.3** The sample of the prefilter shall be submitted for testing in its unpleated state.

### **3.5.3 Activated carbon filter**

**3.5.3.1** When organic toxins and non-corrosive volatile organic agents are to be used, an activated carbon filter shall be installed downstream of the exhaust HEPA-filter.

**3.5.3.2** The carbon filter shall be readily accessible for easy servicing, maintenance and replacement (see 3.3.4). A notice clearly stating the type of absorbence filter fitted and the date of installation or service shall be fixed to the front of the cabinet or control panel.

**3.5.3.3** When use is made of carbon filters, the air in the cabinet shall be exhausted to the outside atmosphere through an exhaust duct the discharge end of which shall be above ground level, at least 3 m clear of any building air-intake or window that can open, and away from pedestrian traffic.

## **3.6 Motor blower(s)**

### **3.6.1 Type and control**

#### **3.6.1.1 Variable speed controls**

One or more motor blowers that are fitted to the cabinet shall be directly driven and have variable speed control(s). Variable speed controls shall be accessible to service personnel but not readily accessible to the everyday cabinet user. When more than one blower is fitted, they shall be electrically interlinked.

#### **3.6.1.2 Class II cabinet with separate recirculation and exhaust motor blowers**

If more than one motor blower is used, the motors shall be so electrically controlled that the correct balance is maintained between the recirculating airflow and the exhaust airflow. They shall be so interlocked that the recirculating motor blower cannot operate unless the exhaust blower is operating.

### **3.6.2 Blower rating and performance**

**3.6.2.1** When a class I safety cabinet is tested in accordance with 7.3.1 with a clean HEPA-filter system and an airflow velocity of at least 20 % above the maximum specified inward airflow velocity of 1,0 m/s (see 4.1.3.2), the motor blower(s) shall be capable of maintaining this airflow velocity for at least 2 h.

**3.6.2.2** When a class II safety cabinet is tested in accordance with 7.3.2 with a clean HEPA-filter system and an airflow velocity of at least 20 % above the maximum specified downward airflow velocity in the work space of 0,50 m/s (see 4.2.4.2.1), the motor blower(s) shall be capable of maintaining this airflow velocity for at least 2 h. In addition, when a class II safety cabinet that is fitted with more than one motor blower is tested in accordance with 7.3.2 with a clean HEPA-filter system and an inward airflow of at least 25 % above the minimum specified inward airflow velocity through the work-access aperture of 0,45 m/s, the motor blower(s) shall be capable of maintaining this airflow velocity for at least 2 h.

### 3.7 Exhaust system

3.7.1 The cabinet shall be so constructed that the air contained in it may be

- a) exhausted to the outside atmosphere, by a system that prevents air from flowing back into the cabinet, or
- b) discharged back into the laboratory (class I and class II cabinets only).

3.7.2 If necessary, an additional airtight exhaust duct of a length as short as possible, but not exceeding 3 m, can be used.

3.7.3 If the use of a short duct is not possible, a separate, additional motor blower shall be fitted as near as possible to the outside discharge end of the external exhaust duct, and a thimble type collector (see figure 1) (class I and class II cabinets only) shall be used at the junction between the cabinet duct and the external exhaust duct. The external blower shall be set to ensure excess extraction at all times. The external blower shall be electronically linked to the cabinet to disable the cabinet if the external blower is not operating correctly.

3.7.4 The air extraction system shall be capable of dealing with external wind conditions and duct resistances. Manufacturers shall specify maximum allowable external resistances to airflow.

3.7.5 To prevent air from flowing back into the cabinet, especially when the fan is switched off, the exhaust duct shall be fitted with an automatic anti-blowback system downstream of the exhaust filters. Anti-blowback valves shall be so constructed that the internal components are visible at all times and that the valve seats can be easily inspected and cleaned. Microswitches or other electrical components or controls shall be outside the duct.

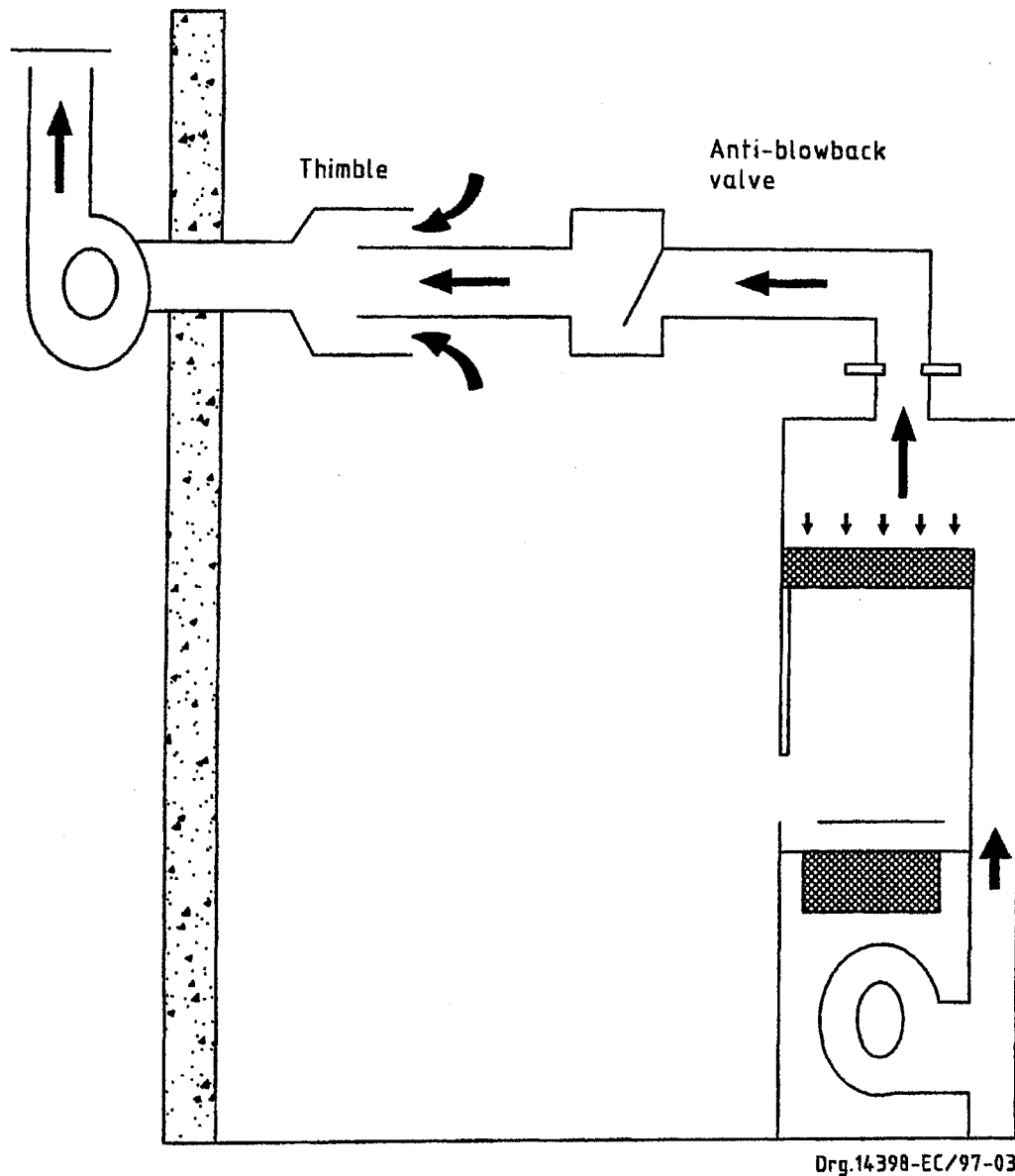
### 3.8 Electrical services

#### 3.8.1 Wiring

Electrical wiring shall be

- a) insulated wiring that complies with SABS 1507:1990, *Electrical cables with extruded solid dielectric insulation for fixed installations (300/500 V to 1 900/3 300 V)*, as published by Government Notice No. 1851 of 1 December 1995 and SABS 1574:1992, *Electrical cables – Flexible cords and flexible cables*, as published by Government Notice No. 1851 of 1 December 1995; or
- b) rubber-insulated wiring that complies with SABS 1520-1:1990, *Flexible electrical trailing cables for use in mines – Part 1: Low-voltage (640/1 100 V and 1 900/3 300 V) cables*, as published by Government Notice No. 1851 of 1 December 1995 and SABS 1520-2:1990, *Flexible electric trailing cables for use in mines – Part 2: High-voltage (3,8/6,6 kV to 19/33 kV) cables*, as published by Government Notice No. 1851 of 1 December 1995, with the said SABS 1574 and with SABS 1576:1993, *Electrical cables – Single core arc welding cable*, as published by Government Notice No. 1851 of 1 December 1995.

Wiring that penetrates boundaries of contaminated areas shall be anchored and their points of entry shall have been made gastight, using non-porous sealants that are not liable to crack or to become porous, or other appropriate means. Electrical components and wiring, other than the blower motor(s) and the associated wiring, shall not be located within the contaminated air zones. All wiring and electrical components within the clean air area of the work space shall be so mechanically secured that no turbulence will be created. Adhesive tape shall not be used for fixing or looming.



**Figure 1 — Example of thimble exhaust system to discharge both cabinet exhaust and laboratory air**

### 3.8.2 Controls

A control enclosure and all controls shall form an integral part of the cabinet. The control enclosure shall contain a control panel. The enclosure shall have a cover that will allow unlimited access to the connections and wiring of the panel. A legible wiring diagram shall be permanently fixed to the cabinet. All operator-adjustable controls shall be clearly visible and easily accessible to the operator. All operation controls and electrical components within the control enclosure shall be identified by being suitably marked in a permanent manner.

### 3.9 Noise levels

When a cabinet is tested in accordance with 6.7, the noise emitted by the cabinet during operation shall not exceed 65 dB(A).

### **3.10 Vibration**

When a cabinet is tested in accordance with 6.3, during the entire operation of the cabinet, the vibration velocity in any plane of any work surface shall have an r.m.s. value of 10 Hz  $\pm$  1 Hz to 250 Hz  $\pm$  25 Hz, not exceeding 0,7 mm/s.

## **4 Specific requirements**

### **4.1 Class I cabinets**

#### **4.1.1 General**

**4.1.1.1** A class I cabinet shall be a self-contained unit that includes at least a work space, prefilters, HEPA-filters and a blower for HEPA-filtered exhaust air. If handling of organic toxins and non-corrosive volatile organic agents is required, an activated carbon filter shall also be included.

**4.1.1.2** The cabinet shall be an independent operating unit and shall be independent of any other air-circulation system.

**4.1.1.3** The exhaust outlet may face in any direction, provided that it is readily accessible.

**4.1.1.4** The work face of the work space shall include a viewing window and a work-access aperture through which an inward flow of air is maintained.

**4.1.1.5** In order to contain potentially hazardous materials within the cabinet, all contaminated zones under positive air pressure shall be surrounded by zones maintained under negative pressure relative to the pressure in the work room/environment.

**4.1.1.6** When a cabinet is tested in accordance with 6.8, the cabinet shall offer a protection factor of at least  $1 \times 10^5$ .

**4.1.1.7** Under no circumstances shall a class I cabinet be upgraded to comply with the requirements of a class III cabinet.

#### **4.1.2 Work floor**

The work floor shall be rigid, flat and constructed in one piece with radiused corners to facilitate cleaning and disinfection (see 3.4.2). The front edge of the floor shall have a retaining lip of height at least 10 mm that serves to contain spillage of liquid within the cabinet.

#### **4.1.3 Flow and distribution of air**

**4.1.3.1** When a smoke test is carried out in accordance with 6.5.3.3, it shall demonstrate that the direction of airflow is inwards over the entire area of the work-access aperture.

**4.1.3.2** When determined in accordance with 6.6.3.1, the average velocity of the air flowing into the work-access aperture shall be in the range 0,7 m/s to 1,0 m/s. No single velocity reading shall differ from the average velocity by more than 20 %.

### **4.2 Class II cabinets**

#### **4.2.1 General**

**4.2.1.1** A class II cabinet shall be a self-contained unit that includes at least a work space, prefilters, HEPA-filters and a blower for unidirectional (laminar) HEPA-filtered airflow and HEPA-filtered exhaust air. If handling of organic toxins and non-corrosive volatile organic agents is required, an activated carbon filter shall also be included.

**4.2.1.2** The cabinet shall be an independent operating unit, and shall be independent of any other air-circulation system.

**4.2.1.3** The exhaust outlet may face in any direction, provided that it is readily accessible.

**4.2.1.4** The work face of the work space shall include a viewing window and a work-access aperture through which an inward flow of air is maintained.

**4.2.1.5** In order to contain potentially hazardous materials within the cabinet, all contaminated zones under positive air pressure shall be surrounded by zones maintained under a negative pressure relative to the pressure in the work room/environment.

**4.2.1.6** The delivery area for the air to the work space shall be free from interposed projections or cavities that could interfere with the containment performance.

**4.2.1.7** When a class II cabinet is tested in accordance with 6.5.3.1, the DOP aerosol penetration at all construction joints bordering the work space shall not exceed 0,03 %.

#### **4.2.2 Work floor**

The work floor shall be firm, shall not be fastened and shall be readily raised but shall have a location-fixing and position-fixing system, as well as systems to prevent reversed installation. The work floor may be solid or perforated. If the work floor is solid, it shall have a retaining lip around its perimeter of height at least 10 mm, that serves to contain spillage of liquid within the cabinet. All corners of the floor shall be radiused to facilitate cleaning and disinfection (see 3.4.2). Components or attachments associated with the work floor in the work space shall be so constructed as to facilitate easy and effective cleaning and disinfection.

#### **4.2.3 Sump**

The sump, which provides the base of the lower air plenum, shall be watertight and all joints shall be welded, ground flush and dressed. The sump shall be sized to retain fluid to a depth of at least 10 mm. The floor of the sump shall be free from obstructions and attachments, and have all corners radiused to facilitate cleaning and disinfection (see 3.4.2). The sides of the lower air plenum (i.e. the area below support structures for the work floor) shall be free from cracks, crevices and sharp projections that could adversely affect cleaning and disinfection.

#### **4.2.4 Flow and distribution of air**

##### **4.2.4.1 Recirculating air and air barrier**

Air shall be recirculated through the work space through HEPA-filters and in a unidirectional (laminar) manner, thus providing contamination-free air for product protection.

An air barrier between the work space and the room shall be created across the full width of the work-access aperture by the induction of atmospheric (room) air downwards into the sump.

##### **4.2.4.2 Velocity and uniformity of airflow in the work space**

**4.2.4.2.1** When determined in accordance with 6.6.3.2, the average velocity of the unidirectional (laminar) flow of air shall be not less than 0,45 m/s and not more than 0,50 m/s. No single velocity reading shall differ from the average velocity by more than 20 %.

**4.2.4.2.2** When a cabinet is tested in accordance with 6.10, the total number of colonies of the test organism, counted after incubation, shall not exceed five in any of the six replicate tests, thus indicating a minimum of cross-contamination and, therefore, an acceptable level of uniformity of airflow.

##### **4.2.4.3 Air barrier integrity**

**4.2.4.3.1** When a cabinet is tested in accordance with both 6.5 and 6.10, the DOP aerosol penetration shall not exceed 0,03 %, and the number of colonies of the test organism counted on the plates shall not exceed five in any test.



**4.2.4.3.2** When a cabinet is tested in accordance with 6.8, the cabinet shall offer a protection factor of at least  $1,0 \times 10^5$ .

**4.2.4.3.3** The mean inward airflow velocity at the work-access aperture shall be at least 0,4 m/s when indirectly measured as exhaust airflow velocity in accordance with 6.6.3.2.2. This shall be achieved at the minimum specified downward airflow velocity of 0,45 m/s in the work space.

**4.2.4.3.4** When a smoke test is carried out in accordance with 6.5.3.3, it shall indicate that the direction of airflow is inwards over the whole area of the work-access aperture.

NOTE – The proportional adjustment of the quantities of barrier air and the work space air is critical to the performance of the cabinet, as is the unidirectional (laminar) flow of the air within the cabinet.

**4.2.4.3.5** When a smoke test is carried out in accordance with 6.5.3.3.2, it shall visually indicate that there is no escape of smoke to the ambient side over the whole area of the work-access aperture. In addition, there shall be no undue turbulence which could lead to backstreaming along the inside of the window.

#### **4.2.4.4 Temperature**

The temperature, measured inside the cabinet at a height of 100 mm above the centre of the work space, shall not rise by more than 8 °C above the ambient temperature in the laboratory after 4 h of continuous working of the motor blower (see 3.6) and the lights turned on.

### **4.3 Class III cabinets**

#### **4.3.1 General**

**4.3.1.1** A class III cabinet shall be a self-contained unit that includes at least a work space, prefilters, HEPA-filters and a blower for HEPA-filtered inlet and exhaust air. Provision shall be made to prevent the backward flow of contaminated air through the air-intake by the fitting of an inlet HEPA-filter that also provides a supply of sterile air to flush the interior and prevent contamination of the material being handled. If handling of organic toxins and non-corrosive volatile organic agents is required, an activated carbon filter shall also be included in the exhaust duct. Exhaust air shall be ducted to the outside atmosphere.

**4.3.1.2** The cabinet shall be an independent operating unit and shall be independent of any other air-circulation system.

**4.3.1.3** When a cabinet is tested in accordance with 6.1.1, it shall be gastight. At no location shall a gas leak in excess of 16,5 g per annum be detected.

**4.3.1.4** The exhaust outlet may face in any direction, provided that it is readily accessible.

**4.3.1.5** The work face of the work space shall include a viewing window and a sealed barrier that separates the operator from the work space. This barrier shall be fitted with gloves that are continuous with the barrier and the outer shell of the cabinet. The gloves shall enable the worker to handle materials inside the cabinet.

**4.3.1.6** All controls associated with the cabinet shall be operated from outside the cabinet.

**4.3.1.7** In order to contain potentially hazardous materials within the cabinet, the interior of the cabinet shall always remain under a negative pressure relative to the pressure in the work room/environment. A manometer with a range of –500 Pa to 500 Pa shall be mounted outside the cabinet to give a visual indication of the pressure of the interior negative pressure plenum.

**4.3.1.8** Any contaminated zone under positive pressure shall be surrounded by zones maintained at negative pressure equal to that maintained in the cabinet work space.

**4.3.1.9** There shall be specific access points in the exhaust duct for the measurement of the airflow rate.

**4.3.1.10** Under no circumstances shall a class I cabinet be upgraded to comply with the requirements of a class III cabinet.

#### **4.3.2 Glove ports**

Manipulation in the work space shall be carried out by means of glove ports which may also serve as transfer ports and for the attachment of transfer bags.

##### **4.3.2.1 Glove port assembly**

**4.3.2.1.1** The glove port assembly shall be attached to the front panel of the cabinet either by means of suitable fasteners with sealing gaskets or shall be permanently welded to, riveted to or pressed from the front panel of the cabinet.

**4.3.2.1.2** The glove port assembly shall comply with all the physical and chemical requirements for the outer shell of the cabinet, as specified in 3.3.1.

**4.3.2.1.3** The dimensions of the glove port assembly shall be such as to provide for the attachment of standard, commercially available beaded glovebox gauntlets, without undue tension on the rims of the gloves. The manufacturer shall specify the glove cuff diameter or shape appropriate to the particular port size.

**4.3.2.1.4** The outer side of the port ring shall have two grooves to accommodate the beaded cuff of the glove and of a secondary glove to permit changing gloves without compromising the seal.

**4.3.2.1.5** A bung that can be fitted internally or externally to provide an efficient and absolute seal of the port shall be provided for each port.

##### **4.3.2.2 Gloves (gauntlets)**

**4.3.2.2.1** The gloves shall fit either hand equally well and shall have beaded cuffs that are compatible with the diameter and shape of the glove ports.

NOTE – Gloves made from translucent material might be affected by DOP aerosol and other aerosols, and therefore steps should be taken to prevent such gloves from coming into contact with such aerosols when filter integrity tests are being conducted.

**4.3.2.2.2** Gloves shall be made of translucent material in order to readily detect any damage to the glove.

**4.3.2.2.3** The gloves shall be easily replaceable from outside the cabinet, by pushing the old glove to the inside of the cabinet and fitting a new glove while the blower is still running.

**4.3.2.2.4** Gloves shall be attached to the ports in such a manner that they do not detach easily when in use. The means of attachment shall not compromise the integrity of the glove.

#### **4.3.3 Filters**

Both the inlet and the exhaust filters of a class III cabinet shall be HEPA-filters of a size appropriate for handling at least the specified airflow (see 4.3.4.2), and they shall be of size and specification given in 3.5.1.

#### **4.3.4 Flow and distribution of air**

**4.3.4.1** When determined in accordance with 6.6.3.3.1, the airflow velocity through the glove ports, when all gloves are detached, shall be at least 0,75 m/s.

**4.3.4.2** When determined in accordance with 6.6.3.3.2, the airflow through the inlet filter, when the gloves are attached, shall be at least 3 m<sup>3</sup>/min.

#### 4.3.5 Work floor

The work floor shall be rigid, flat and constructed in one piece with radiused corners to facilitate cleaning and disinfection (see 3.4.2).

#### 4.3.6. Transfer chamber

**4.3.6.1** A transfer chamber may be fitted to the cabinet to permit the transfer of bulky items into the cabinet. If fitted, the transfer chamber shall be of a suitable size, with doors appropriate to the size of the items in question, and shall be fitted to the side of the cabinet.

**4.3.6.2** The transfer chamber shall be a seamless gastight one-piece chamber with radiused corners to facilitate cleaning and disinfection. When a transfer chamber fitted to the cabinet is tested in accordance with 6.1, the seams and joints of the doors and of the chamber shall show no sign of gas leakage. All materials used for the construction of the chamber shall comply with the requirements of 3.3.1. The chamber shall be fitted with in-line HEPA-filters and suitable valves to allow partial evacuation of the chamber, when required.

### 5 Transport and installation of cabinets

#### 5.1 Transport

The cabinet and its components shall be so transported and installed that damage to any part of the cabinet is prevented and the integrity of the cabinet is ensured.

#### 5.2 Installation

After installation, the following tests shall be performed to ensure that the cabinet complies with the relevant performance and safety requirements.

##### a) Gastightness of the outer shell (class III cabinets)

When tested in accordance with 6.1, a class III cabinet shall comply with the requirements of 4.3.1.3.

##### b) HEPA-filter and HEPA-filter installation integrity

When tested in accordance with 6.4, class I, class II and class III cabinets shall comply with the requirements of 3.5.1.3.1.

##### c) Integrity of the viewing window seal

When tested in accordance with 6.5, a class II cabinet shall comply with the requirements of 3.4.3.4.

##### d) Flow and distribution of air, average velocity and uniformity and rate of airflow

**Class I cabinets:** When determined in accordance with 6.6.3.1, the flow and distribution of air through the work-access aperture shall comply with the requirements of 4.1.3.2.

When a smoke test is carried out in accordance with 6.5.3.3, the airflow shall comply with the requirements of 4.1.3.1.

**Class II cabinets:** When determined in accordance with 6.6.3.2.1, the velocity and uniformity of airflow in the work space shall comply with the requirements of 4.2.4.2.1.

When determined in accordance with 6.6.3.2.2, the inward airflow velocity through the work-access aperture shall comply with the requirements of 4.2.4.3.3.

**Class III cabinets:** When determined in accordance with 6.6.3.3, the airflow shall comply with the requirements of 4.3.4.1 and 4.3.4.2.

## **6 Methods of test**

### **6.1 Determination of gastightness of outer shell (class III cabinets)**

#### **6.1.1 Principle**

The cabinet is sealed and positively pressurized with hydrofluorocarbon gas. All surfaces and joints are scanned with the detector probe for leakage of the gas.

#### **6.1.2 Apparatus**

**6.1.2.1 Gas detector**, adjusted and calibrated to detect, at a reference leak source, the loss of hydrofluorocarbon gas at a maximum rate of 16,5 g per annum.

**6.1.2.2 Manometer**, with scale divisions not exceeding 10 Pa and that is capable of registering pressures in the range 200 Pa to 300 Pa.

**6.1.2.3 Cylinder of hydrofluorocarbon gas (1.1.1.2 tetra-fluoro-ethane)**, commercially available as a refrigerant, R134(a), with a regulator valve, nozzle and connecting hose.

#### **6.1.3 Procedure**

**6.1.3.1** Prepare the cabinet for testing as a closed system by sealing all openings such as the exhaust opening, removable panel and other penetrations by any convenient means. Remove all external covers that are not essential for the operation of the cabinet.

**6.1.3.2** Attach the manometer to the relevant test area of the cabinet to indicate interior pressure.

**6.1.3.3** Suitably connect the gas cylinder to the test area and release the gas to positively pressurize the cabinet interior to a pressure of 250 Pa  $\pm$  10 Pa.

**6.1.3.4** Prepare, calibrate and operate the gas detector in accordance with the manufacturer's instructions.

**6.1.3.5** Move the probe of the instrument over the seams, joints, utility penetrations, gaskets and other locations of possible leakage, keeping the probe 7 mm to 12 mm from any surface and moving it at a rate of approximately 0,013 m/s.

#### **6.1.4 Evaluation**

Deem the cabinet to be gastight if at no location a gas leak in excess of 16,5 g per annum is detected.

### **6.2 Determination of illuminance**

#### **6.2.1 Principle**

Measurements of illuminance are taken at evenly spaced locations at a specified work level.

#### **6.2.2 Apparatus**

**Illuminance meter** (calibrated, cosine and vision-corrected), of such range that the illuminance measured is at least one-fifth of the full-scale value.

### 6.2.3 Procedure

6.2.3.1 Operate the lamps in the cabinet for at least 2 h.

6.2.3.2 Take eight illuminance measurements at eight evenly spaced locations at a height not exceeding 25 mm from the surface of the work floor but not within 150 mm of the perimeter of the work space. Record the results obtained at each location.

## 6.3 Determination of vibration

### 6.3.1 Principle

Measurements of the vibration velocity are made with a simple vibration meter at the geometric centre of the work surface, both with and without the cabinet in operation, to permit comparison of the vibration levels under these two conditions. Determination of the net vibration, i.e. vibration attributable to the cabinet alone, would require vibration frequency analysis.

### 6.3.2 Apparatus

**Vibration meter**, capable of measuring steady-state vibration velocities in the range 0,05 mm/s-1,0 mm/s (r.m.s.) in the frequency range 10 Hz  $\pm$  1 Hz to 250 Hz  $\pm$  25 Hz.

### 6.3.3 Procedure

6.3.3.1 Attach the vibration meter to the geometric centre of the work surface.

6.3.3.2 Ensure that the airflow is as specified.

6.3.3.3 With the cabinet in normal operation, measure the gross vibration velocity in the vertical, horizontal front to rear, and horizontal side-to-side axes.

6.3.3.4 Turn off the mechanical system and with the sensing element positioned and attached as in 6.3.3.1, measure the ambient vibration velocity.

NOTE – The vibration frequency components of the ambient vibration are usually quite different from those of the mechanical cabinet system and hence the derivation of the net r.m.s. velocity (that attributable to the cabinet equipment) from measurements of gross and ambient vibration is not necessarily a simple mathematical subtraction.

### 6.3.4 Report

The following information shall be reported:

- a) maximum value of the gross r.m.s. vibration velocity; and
- b) maximum value of the ambient r.m.s. vibration velocity.

## 6.4 Determination of HEPA-filter and HEPA-filter installation integrity and integrity of gaskets and construction joints in the vicinity of the HEPA-filter installation

### 6.4.1 Principle

A polydisperse aerosol at ambient temperature is fed into the upstream side of the HEPA-filter installation at a specified flow rate and the downstream surface sides of the filters, seals, gaskets and construction joints in the vicinity of the filter installation is scanned with a probe nozzle to determine the percentage of penetration.

## 6.4.2 Apparatus

6.4.2.1 Vane anemometer or thermo-anemometer, as appropriate, accurate to within 2 %.

### 6.4.2.2 DOP generator

6.4.2.2.1 A cold DOP aerosol generator fitted with suitable nozzles and using filtered, compressed air at a pressure of  $140 \text{ kPa} \pm 14 \text{ kPa}$ , with the free airflow adjusted to not less than  $30 \text{ l/min}$  per nozzle, producing an aerosol of particles with a median diameter of less than  $0,8 \text{ }\mu\text{m}$ .

6.4.2.2.2 A flexible aerosol delivery hose or tubing of nominal internal diameter  $50 \text{ mm}$ .

#### NOTES

1 A hot DOP aerosol generator that uses compressed nitrogen gas may be used to determine the HEPA-filter installation integrity, provided that the aerosol particles produced should comply with the provisions in 6.4.2.2.1.

2 Liquids other than DOP may be used to generate an aerosol of particles, provided that the aerosol generator and the photometer have been suitably adjusted and calibrated for the alternative liquid. The aerosol produced should have a similar particle size distribution to that given in 6.4.2.2.1.

### 6.4.2.3 Aerosol photometer

A light-scattering mass concentration indicator fitted with a probe nozzle. The probe nozzle or tip used for filter integrity testing is of internal diameter ( $d$ ) not exceeding  $30 \text{ mm}$ . Any transition from initial inlet diameter to final inlet diameter is gradual. A maximum excluded angle  $\theta$  of  $20^\circ$  is recommended. Photometers that have a threshold sensitivity of at least  $10^{-3} \text{ }\mu\text{g/l}$  for DOP particles of diameter  $0,3 \text{ }\mu\text{m}$ , and that are capable of measuring concentrations in the range of  $80 \text{ }\mu\text{g/l}$  to  $120 \text{ }\mu\text{g/l}$  are suitable. The test photometer has a sample flow rate of  $30 \text{ l/min} \pm 3 \text{ l/min}$ . The probe inlet is of sufficient size to maintain the probe inlet rate at or slightly higher than a test flow rate of  $27,5 \text{ l/min}$  through the filter.

## 6.4.3 Procedure

NOTE – The test operator should avoid inhalation and exposure to heavy concentrations of the test aerosol. It is recommended that a suitable mask or respirator be worn for the duration of the test and thereafter, if necessary.

6.4.3.1 By using the method given in 6.6, determine the airflow and ensure that the flow through the air filter bank is within the operating limits of the cabinet design flow (see 4.1.3.2, 4.2.4.2, 4.3.4.1, or 4.3.4.2, as applicable). Ensure that the cabinet is operating normally while this procedure is being carried out.

6.4.3.2 Regulate the generator pressure and the gas flow rate in accordance with the manufacturer's instructions or as specified in 6.4.2.2.1, as appropriate.

Introduce the aerosol via a sparge arrangement, if necessary, so that it is evenly distributed across the air entry.

### 6.4.3.3 For photometers that have

- a) a linear readout, establish the upstream concentration by introducing the least amount of DOP aerosol required to produce a 100 % reading, thus allowing the instrument to be straylight adjusted to zero on the lowest scale range when the sample air stream is filtered free of aerosol, and
- b) a logarithmic readout, adjust the upstream concentration (as determined from the instrument calibration curve) by introducing the least amount of DOP aerosol required to produce a concentration of  $1 \times 10^4$  above that concentration required to give a reading of one scale division. Avoid prolonged exposure of filters to DOP.

6.4.3.4 With any removable filter guard removed, scan the entire filter media face in slightly overlapping strokes at a distance between the probe and the filter media face of approximately  $25 \text{ mm}$  and at a traverse rate not exceeding  $50 \text{ mm/s}$ . Ensure that each filter pleat is scanned parallel to the

direction of the pleat. Also scan the entire periphery of the filter at the bond between the filter media and the frame, at the seal between the filter frame and the cabinet, and any construction joints downstream of the filter installation. Record any local areas or points where a reading exceeding 0,03 % is obtained.

## 6.5 Method for the detection of leaks into the work space of, and demonstration of the integrity of a class II cabinet and its air barrier

### 6.5.1 Principle

6.5.1.1 While air-generated DOP aerosol is directed at joints in the vicinity of the work space or at the work-access aperture (air barrier), measurements are made using an aerosol photometer.

Any meter readings in excess of 0,03 % penetration are an indication of seal or joint leakage or induction of contaminants into the clean work zone.

6.5.1.2 Smoke is released on the work space side of the work-access aperture.

Escape of smoke to the ambient air indicates an ineffective air barrier.

### 6.5.2 Apparatus

6.5.2.1 DOP generator, as in 6.4.2.2.

6.5.2.2 Aerosol photometer, as in 6.4.2.3.

6.5.2.3 Air current tube (smoke generating tube).

6.5.2.4 **Barrier test fitting.** (For barrier integrity testing, the aerosol delivery hose is fitted with a smooth parallel-bore fitting of internal diameter  $50 \text{ mm} \pm 1 \text{ mm}$  at the point of discharge. The fitting is of overall length  $250 \text{ mm} \pm 5 \text{ mm}$ , incorporates flow straighteners at its inlet and has a square cut end as illustrated in figure 2.)

Dimensions in millimeters

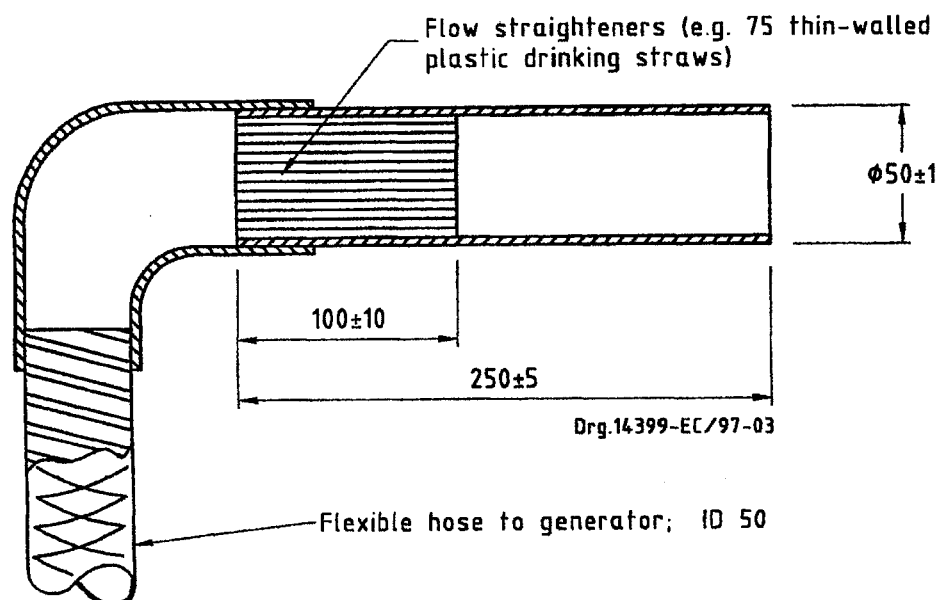


Figure 2 — Barrier test fitting

### 6.5.3 Procedure

NOTE – The test operator should avoid inhalation and exposure to heavy concentrations of the test aerosol. It is recommended that a suitable mask or respirator be worn for the duration of the test and thereafter, if necessary.

#### 6.5.3.1 Joints and seals

**6.5.3.1.1** Using the aerosol photometer, measure the ambient aerosol level of the work room/environment and of the work space of the cabinet. If the reading of the aerosol photometer is less than  $10^3$  above the filter face reading, discharge sufficient aerosol at the exterior of the joint or seal from a distance of approximately 150 mm to ensure the challenge is maintained at 0,1 % concentration or more, with the photometer setting as that used to establish the 100 % baseline during the integrity testing of the HEPA-filter.

**6.5.3.1.2** Use the photometer to scan all construction joints bordering the work space. Hold the probe nozzle inside the cabinet, not more than 25 mm away from the joint and move it along the joint at not more than 5 cm/s (see figure 3).

**6.5.3.1.3** Start scanning approximately 3 s after the aerosol cloud has been directed at the joint.

Dimensions in millimeters

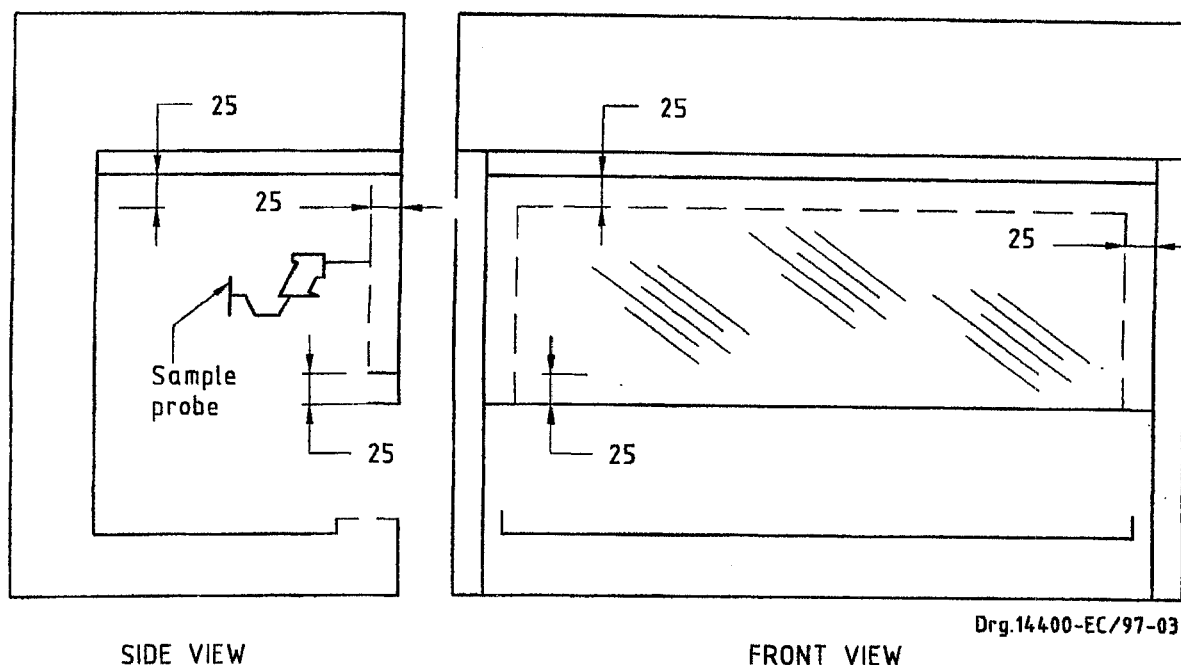


Figure 3 — Scanning of joints and seals

#### 6.5.3.2 Air barrier

**6.5.3.2.1** Discharge sufficient cold aerosol at a distance of approximately 150 mm in front of each test position and scan the lower edge of the viewing glass with the probe inlet held inside the work space at a distance of 25 mm away from the glass and at 100 mm centres. Start scanning at a point 25 mm from each of the work surface boundaries (see figure 4).

**6.5.3.2.2** Direct the probe inlet towards the work-access aperture and position its centre approximately 10 mm above the lower edge of the viewing window.

**6.5.3.2.3** Operate the aerosol photometer at each test position for at least 15 s. Where an intermittent penetration reading greater than 0,03 % above the ambient concentration is obtained, continue for at least a further 30 s.

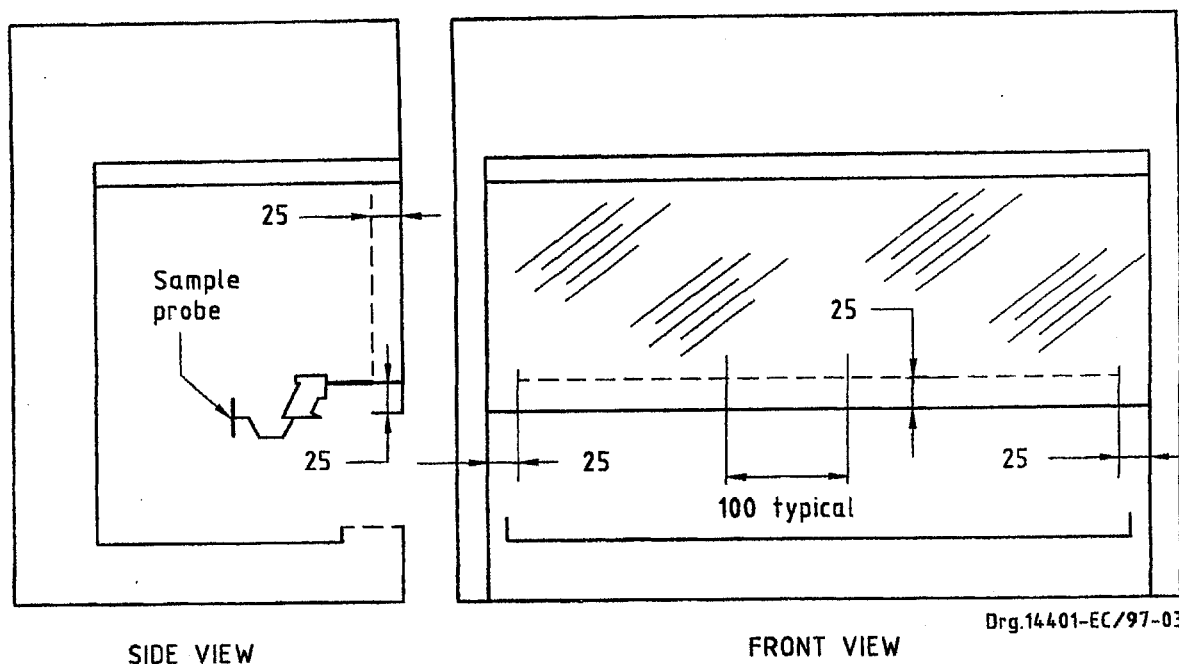
**6.5.3.2.4** Scan the front edge of the work floor (not the front edge of the cabinet) at a distance of 25 mm and at 100 mm centres. Commence scanning at a point 25 mm from each of the work surface boundaries (see figure 5). Direct the probe inlet towards the work-access aperture and ensure that its



centre is positioned approximately 25 mm above the work floor.

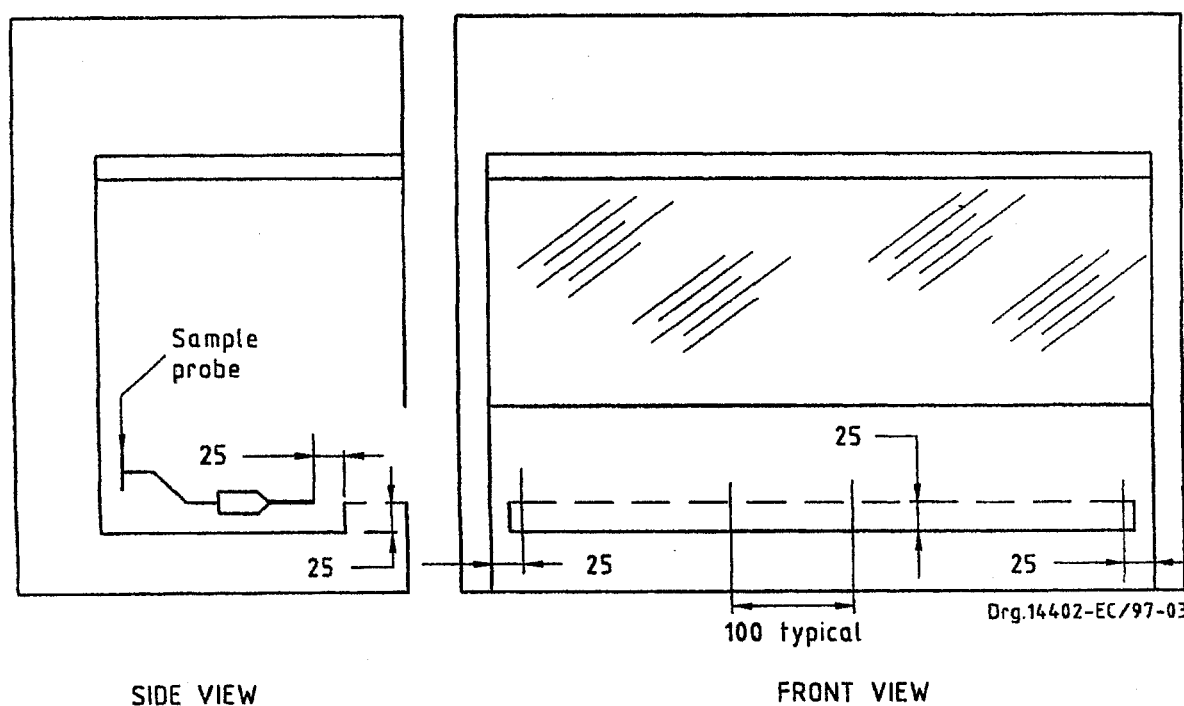
**6.5.3.2.5** Operate the photometer at each test position for at least 15 s and record any photometer reading, and its location, in excess of 0,03 % aerosol penetration, relative to the 100 % measured upstream. Where an intermittent penetration reading greater than 0,03 % above the ambient concentration is obtained, continue for at least a further 30 s.

Dimensions in millimeters



**Figure 4 — Scanning of the top edge of the work-access aperture**

Dimensions in millimeters



**Figure 5 — Scanning of the front edge of the work floor****6.5.3.3 Smoke test**

**6.5.3.3.1** In addition, carry out a simple smoke test to determine the direction of airflow near the work-access aperture. Generate aerosol or any other smoke on the ambient side of the aperture so that the smoke cloud is within 150 mm of the entire area of the opening and note the direction of the airflow.

**6.5.3.3.2** Using the air current tube (smoke generating tube) (see 6.5.2.3), release an even stream of smoke at a distance of 70 mm  $\pm$  5 mm from the inside plane of the work-access aperture at a series of positions with the tip of the air current tube approximately 25 mm below the bottom edge of the viewing window. Test positions at approximately 50 mm from both inner side walls of the work space of the cabinet, and at intervals of approximately 100 mm between these points.

**6.6 Determination of flow and distribution of air, average velocity and uniformity and rate of airflow****6.6.1 Principle**

Airflow velocity readings are taken at selected locations, using an anemometer to determine the average airflow velocity, the uniformity of airflow and the airflow rate.

**6.6.2 Apparatus**

**6.6.2.1 Vane anemometer**, free-standing, where applicable, of appropriate vane diameter and accurate to within  $\pm$  2 %.

**6.6.2.2 Thermo-anemometer**, where applicable, and accurate to within  $\pm$  2 %.

**6.6.2.3 Barometer.**

**6.6.2.4 Thermometer.**

**6.6.3 Procedure****6.6.3.1 Class I cabinets**

Ensure that the cabinet is operating normally. With the vane anemometer in the plane of the work-access aperture, take and record velocity readings of the air flowing into the aperture for at least 1 min and at at least five locations evenly distributed across the plane of the work-access aperture.

**6.6.3.2 Class II cabinets****6.6.3.2.1 Downflow**

**6.6.3.2.1.1** Ensure that the cabinet is operating normally. Take and record velocity readings in the horizontal plane 100 mm above the top edge of the work-access aperture using a free-standing vane anemometer.

**6.6.3.2.1.2** Take velocity readings at 200 mm to 225 mm intervals in both directions, starting at a location 75 mm to 100 mm from the inner edge of the work surface. Record each reading and its location.

**6.6.3.2.1.3** Record the pressure drop across the filter system as indicated by the manometer or gauge fitted to the cabinet.

**6.6.3.2.2 Inflow**

**6.6.3.2.2.1** Ensure that the cabinet is operating normally.

**6.6.3.2.2.2** Using a suitable vane anemometer or thermo-anemometer (as applicable), take readings at multiple points on a plane bounded by the perimeter of the exhaust aperture or duct. Record each

reading and calculate the mean airflow velocity.

**6.6.3.2.2.3** Multiply the area of the exhaust aperture or duct by the mean velocity obtained to yield the volumetric discharge rate of effluent air.

**6.6.3.2.2.4** Obtain the average inward airflow velocity at the work-access aperture by dividing the volumetric effluent air volume by the cross-sectional area of the work-access aperture.

### **6.6.3.3 Class III cabinets**

#### **6.6.3.3.1 Inflow through open glove ports**

**6.6.3.3.1.1** Ensure that the cabinet is operating normally.

**6.6.3.3.1.2** Remove the gloves. Using the vane anemometer placed at the centre of each open glove port, take and record the airflow velocity for at least 1 min.

#### **6.6.3.3.2 Inflow through the inlet filter**

**6.6.3.3.2.1** Ensure that the cabinet is operating normally with the gloves attached.

**6.6.3.3.2.2** Using the thermo-anemometer, take multiple measurements within the exhaust duct along two axes perpendicular to each other, and record the mean airflow velocity.

**6.6.3.3.2.3** Multiply the area of the exhaust duct by the average velocity obtained to yield the volumetric discharge rate of effluent air which is equal to the inflow through the inlet filter.

### **6.6.4 Report**

#### **6.6.4.1 Class I**

Report

- a) each velocity reading and its location;
- b) the average of the velocity readings taken;
- c) maximum and minimum velocity readings; and
- d) percentage variations from the average of the maximum and minimum readings.

#### **6.6.4.2 Class II**

##### **6.6.4.2.1 Downflow**

Report

- a) the pressure drop across the filter system;
- b) each velocity reading and its location;
- c) the average of the velocity readings taken;
- d) maximum and minimum velocity readings; and
- e) percentage variations from the average of the maximum and minimum readings.

##### **6.6.4.2.2 Inflow**

Report

- a) the pressure drop across the exhaust filter system;
- b) exhaust airflow velocity (m/s);
- c) dimensions of the exhaust duct and work-access aperture; and

d) mean inward airflow velocity (m/s).

#### 6.6.4.3 Class III

Report

- a) inflow velocity, right glove port (m/s);
- b) inflow velocity, left glove port (m/s);
- c) exhaust airflow velocity (m/s);
- d) dimensions of exhaust duct; and
- e) inlet airflow rate (m<sup>3</sup>/min).

### 6.7 Determination of noise level

#### 6.7.1 Principle

Noise levels are measured at selected locations near the cabinet under normal operating conditions and the background ambient conditions are also recorded.

#### 6.7.2 Apparatus

##### 6.7.2.1 Sound level meter

Use an integrating sound level meter configuration, that complies at least with the accuracy requirements specified for a type 1 instrument in SABS IEC 60651:1979, *Sound level meters*, and SABS IEC 60804:1985, *Integrating-averaging sound level meters*, as published by Government Notice No. 399 of 1 April 1999. Use a windscreen of a type specified by the sound level meter's manufacturer as being suitable for the particular microphone and that does not detectably influence the accuracy of the meter under the ambient conditions of the test.

NOTE – In principle, no time weighting other than I-time weighting is allowed during integration: S-time weighting in particular should be disabled when  $L_{Aeq,T}$  is measured since it could introduce errors over short integration intervals.

##### 6.7.2.2 Calibration source

As the calibration source, use a sound calibrator that complies with the requirements prescribed for a type 1 calibrator in SABS IEC 60942:1997, *Electroacoustics – Sound calibrators*, as published by Government Notice No. 399 of 1 April 1999.

#### 6.7.3 Procedure

**6.7.3.1** Ensure that the cabinet is operating normally. For class I and class II cabinets, measure and record the noise level with the sound level meter situated 0,3 m from, and 0,3 m above, the top edge of the work-access aperture, at the vertical centre line of the cabinet and 1 m from any other part of the cabinet including the duct work and from the discharge point of the extraction system, if fitted. For class III cabinets, take measurement with the sound level meter situated 0,6 m above the work surface of the cabinet and 0,3 m from the cabinet front, at the vertical centre line of the cabinet and 1 m from any duct work and from the discharge point of the extraction system, if fitted.

**6.7.3.2** Ensure that the airflow of the cabinet is as specified. Take all measurements with the sound level meter set to use the A-weighted network and fast response. Using the acoustic calibration source, check the acoustic sensitivity of the sound level meter before and immediately after the measurements are made and discard the results if the two checks do not coincide to within 1,0 dB.

#### 6.7.4 Report

Include the following details in the report:

- a) all operating noise level measurements and their location;
- b) the identified maximum noise level and its location; and

c) the ambient noise level measurements at locations where indicated.

## 6.8 Determination of the protection factor for class I and class II cabinets

### 6.8.1 Principle

**6.8.1.1** Tests which are performed to assure that aerosols will be contained within open-fronted microbiological safety cabinets, are specified in terms of an operator protection factor. This factor expresses the leakage from the open front of a cabinet, of a given aerosol that was released within the work space.

**6.8.1.2** The transfer index defines the exposure experienced at a given point as a result of the release of a known amount of tracer substance (bacterial spores or potassium iodide particles), within the cabinet. This exposure is defined as  $n/(Ns)$ , where  $N$  is the number of particles released and  $n$  is the number of particles recovered at a sampling rate of  $s$ , the sampling being continued to completion. The transfer ratio index with and without the cabinet defines the protection factor and it is necessary to define the reference situations which represent the open bench conditions, that is the exposure that an operator is subjected to by working in a ventilated room without the use of a safety cabinet. The reference open-bench conditions are defined as a room with a ventilation rate  $V$ , of  $10 \text{ m}^3/\text{min}$ , with complete mixing. The transfer index of the reference room is equal to  $1/V = 1/10$  and the protection factor then becomes  $(Ns)/(10n)$  if the sampling rate  $s$  is expressed in cubic metres per minute, or  $(Ns)/(10^4n)$ , if  $s$  is expressed in litres per minute. The minimum value of the protection factor that can be determined depends on the sensitivity and selectivity of the test, i.e. the magnitude of the challenge,  $N$ , the sampling rate  $s$ , and the least number of particles recovered that can be readily distinguished from background contamination. Practical values for these are  $N$  at least  $3 \times 10^6$ ,  $s$  at least  $50 \text{ l/min}$  and  $n$  not exceeding 10, which leads to a minimum verifiable value of not less than  $1,5 \times 10^5$  for the protection factor.

**6.8.1.3** An "artificial arm" in the form of a cylinder of diameter between 60 mm and 65 mm, is used to mimic the turbulence produced by the worker's arm at the front aperture.

**6.8.1.4** Containment tests on safety cabinets can be performed

- a) with a microbiological aerosol that consists of a fine spray of microorganisms produced by a nebulizer charged from an aqueous suspension, or
- b) with an airborne challenge of potassium iodide particles produced by a spinning disk aerosol generator.

### 6.8.2 Microbiological method

NOTE – In a room where cross-contamination, external contamination or protection factor tests have recently resulted in considerable leakage of the bacterial challenge into the ambient air, it is particularly advisable to perform a background test for the presence of the test organism 24 h before performing protection factor tests. A count of more than five test organisms on one of the culture plates following a 10 min test should be regarded as unsatisfactory, and the protection factor tests should be postponed until the ambient air is no longer contaminated with the test organisms. It is advisable to perform the protection factor tests before the cross-contamination and external contamination tests.

#### 6.8.2.1 Apparatus and materials

##### 6.8.2.1.1 Spore suspension

A suspension of spores of a non-pathogenic microorganism, for example *Bacillus subtilis* var. globigii (SABS Type Culture Collection (SABSTCC) Bac 35) in sterile distilled water, standardized to contain approximately  $10^8$  spores to  $10^9$  spores per millilitre.

##### 6.8.2.1.2 Culture plates

Petri dishes of diameter 90 mm that contain 15 ml to 20 ml of nutrient agar (see 6.8.2.1.5).

##### 6.8.2.1.3 Slit air samplers

Two slit air samplers, each being able to operate at between 25 l and 30 l of air per minute.

#### 6.8.2.1.4 Nebulizer

A Collison six-jet nebulizer with an internal outlet of diameter 14 mm, operated from a pressure line at 70 kPa, that sprays approximately 0,2 ml/min and discharges not more than 10 l/min of free air at a velocity of 0,8 m/s.

#### 6.8.2.1.5 Nutrient agar

##### 6.8.2.1.5.1 Ingredients

Agar .....	15,0 g
Peptone .....	10,0 g
Beef extract .....	5,0 g
Sodium chloride .....	5,0 g
Water .....	1 000 ml

##### 6.8.2.1.5.2 Procedure

Dissolve the ingredients in the water by heating. Adjust the pH value to 7,2. Sterilize in bulk by autoclaving at 121 °C for 15 min. Cool to 45 °C and aseptically dispense 15 ml in sterile Petri dishes. Ensure that the surfaces of the Petri dishes are dry before use.

#### 6.8.2.1.6 M1 agar

##### 6.8.2.1.6.1 Ingredients

Nutrient broth .....	3,125 g
Manganese sulphate tetrahydrate .....	0,03 g
Dipotassium hydrogen phosphate .....	0,25 g
Agar (Oxoid no. 3) .....	12,0 g
Water sufficient to produce .....	1 000 ml

##### 6.8.2.1.6.2 Procedure

Dissolve the ingredients in the water by heating. Dispense 30 ml volumes in medical flat bottles or 150 ml volumes in Roux flasks. Sterilize by autoclaving at 121 °C for 15 min. Cool to 45 °C and place the medical flat bottles on a 1-in-4 sloped surface and the Roux flasks on a flat surface. Allow the agar to solidify.

#### 6.8.2.1.7 Cylinder

A cylinder of length approximately 1 m and of diameter 60 mm to 65 mm, that has a smooth surface and is closed at both ends.

#### 6.8.2.2 Preparation of *Bacillus subtilis* var. *globigii* spores

**6.8.2.2.1** Prepare about 20 M1 agar slopes (see 6.8.2.1.6) in medical flat bottles or in Roux flasks, as required, to yield the appropriate amount of spores.

**6.8.2.2.2** Use a fresh culture of *Bacillus subtilis* var. *globigii* that had been subcultured for three days at 36 °C ± 1 °C.

**6.8.2.2.3** Inoculate the agar slopes with the organism and incubate for one week at 36 °C ± 1 °C and then at room temperature until 80 % sporulation has been obtained (usually within about 10 d).

**6.8.2.2.4** Make a spore stain of the culture after about eight days to determine the percentage of spores. If the percentage is less than 80 %, leave the cultures until 80 % sporulation has been obtained.

NOTE – Use a 5 % aqueous malachite green stain for determining the percentage of spores. Make a smear on a glass

microscope slide and heat fix it. Place the slide over a small beaker of boiling water. (Rest the slide on the rim of the beaker.) Add malachite green fire to the slide. Leave on for five minutes, then wash off with water. Counter stain with safranin for approximately 30 s. Rinse off with water. Check under the microscope.

**6.8.2.2.5** Once 80 % sporulation has occurred, very gently wash the culture off the slopes by means of a sterile glass rod and suspend the spores in 10 ml of sterile distilled water. Do not get pieces of agar in the suspension since this will allow the spores to germinate.

**6.8.2.2.6** Centrifuge the suspension in sterile tubes with the tops covered with brown paper. Wash the spores three times with sterile distilled water, i.e. decant the supernatant and add fresh sterile water to remove all traces of the medium. Centrifuge each time for 20 min.

**6.8.2.2.7** After washing, resuspend the spores in sterile water and heat shock at 60 °C for 30 min or at 70 °C for 20 min for three days in succession.

**6.8.2.2.8** Prepare tenfold serial dilutions of spore suspensions and plate out 0,1 ml samples of each dilution on nutrient agar plates (see 6.8.2.1.5) and incubate the plates at 36 °C ± 1 °C for 24 h to 48 h. Count the number of colonies on those plates with between 30 colonies and 300 colonies. From this result, calculate the concentration of spores per millilitre of suspension.

**6.8.2.2.9** Store the stock culture ( $2 \times 10^9$  spores to  $4 \times 10^9$  spores/ml) at 4 °C until needed.

### **6.8.3 Procedure for determining protection factor**

#### **6.8.3.1 General**

For cabinets of width up to 1 m, carry out five replicate protection tests at the centre of the work-access aperture. For cabinets of width exceeding 1 m but not exceeding 1,9 m, carry out five replicate protection tests at the centre of the aperture, and five each at the centres of the right and left halves of the aperture respectively. In order to avoid confusion from background contamination, carry out the tests in a well-ventilated room, after estimating background contamination.

**6.8.3.2** Precede this entire procedure by a control run with the nebulizer switched off.

**6.8.3.3** Introduce the cylinder through the work-access aperture of the cabinet to disturb the airflow (to simulate an operator's arm). Centre the cylinder between the side walls of the safety cabinet work space and, where appropriate, at the centre of the right and left halves of the work access aperture, and normal to the plane of the aperture, extending from the back of the work space to protrude at least 250 mm into the room from the plane of the aperture. Raise the lower surface of the cylinder to between 65 mm and 75 mm from the cabinet floor.

#### **6.8.3.4 Measurement of the concentration of spores in the spore suspension**

**6.8.3.4.1** Prepare tenfold serial dilutions of the stock spore suspension (see 6.8.2.2.9) and plate out 0,1 ml samples of each dilution on nutrient agar plates (see 6.8.2.1.5).

**6.8.3.4.2** Transfer a measured volume of the stock spore suspension (5 ml to 10 ml, as appropriate) into the nebulizer (see 6.8.2.1.4) and weigh the nebulizer with its contents. After spraying, weigh the nebulizer again.

**6.8.3.4.3** Prepare tenfold serial dilutions of the spore suspension that remains in the nebulizer and plate out 0,1 ml samples of each dilution on nutrient agar plates.

**6.8.3.4.4** At least half of the original volume of the stock spore suspension shall remain in the nebulizer. Determine the volume as follows:

$$V + (M_2 - M_1) \geq (V/2)$$

where

$M_1$  is the mass of the nebulizer plus contents before spraying, in grams;

$M_2$  is the mass of the nebulizer plus contents after spraying, in grams; and

$V$  is the volume of the initial spore suspension in the nebulizer, in millilitres;

and assuming the density of the spore suspension to be 1,0 g/ml.

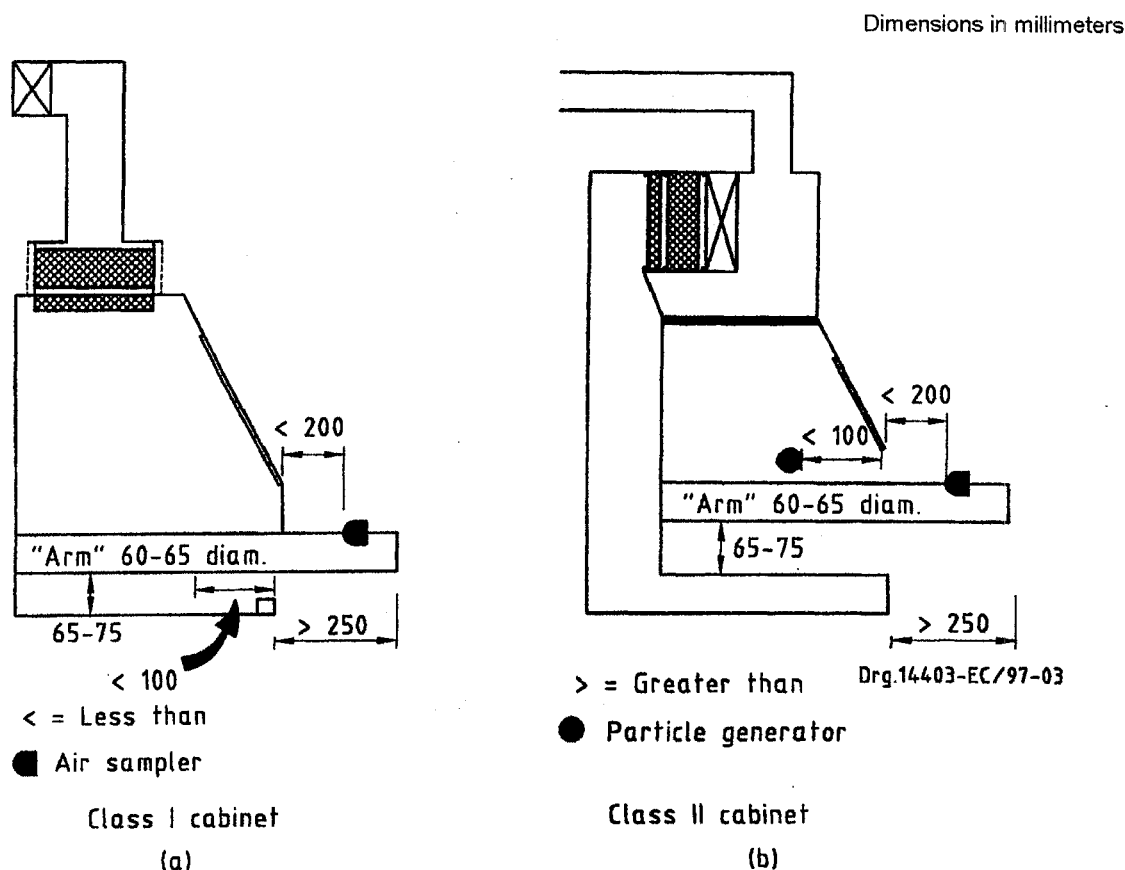
**6.8.3.4.5** Incubate both sets of inoculated plates (before and after spraying) at  $36\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$  for a period of 24 h to 48 h and count the colonies on those plates that have between 30 colonies and 300 colonies on them.

**6.8.3.4.6** From these counts, determine the concentration of spores per millilitre in the initial suspension  $n_1$  and in the final suspension  $n_2$ .

**6.8.3.5** Place the nebulizer inside the work space, and where appropriate, at the centre of the right and left halves of the work-access aperture, with its outlet or the appropriate extension thereof 100 mm behind the plane of the work-access aperture midway between the side walls of the work space, and directed towards the aperture, with the spray axis parallel to the work surface.

**6.8.3.6** For class I cabinets (see figure 6(a)), ensure that the spray axis is below the cylinder and approximately midway between its lower surface and the work surface.

**6.8.3.7** For class II cabinets (see figure 6(b)), ensure that the spray axis is level with the upper edge of the aperture.



**Figure 6 — Diagram that shows the positions of the "artificial arm", the particle generator and the air sampler for testing for operator protection**

**6.8.3.8** Position the slit air samplers outside the cabinet in front of the work-access aperture, with their inlets not more than 200 mm in front of the plane of the aperture. Ensure that the inlets are level with the top of the cylinder, one to the right and one to the left, and each not more than 150 mm from the axis of the cylinder.

**6.8.3.9** Switch on the cabinet and allow it to run until normal operating conditions are reached. Adjust



each sampler to a sampling rate of not less than 25 l/min and not more than 30 l/min. Place a nutrient agar plate (see 6.8.2.1.5) in each slit air sampler. Start the samplers. After 30 s, start the nebulizer that contains the stock spore suspension. Run the nebulizer for a period of not less than 4 min to ensure the dispersal of at least  $3 \times 10^8$  spores. Switch off the nebulizer and continue to run the samplers for a further 5 min.

**6.8.3.10** Incubate the culture plates at  $36^\circ\text{C} \pm 1^\circ\text{C}$  for 24 h to 48 h.

**6.8.3.11** After incubation, count the *Bacillus subtilis* var. globigii colonies. If there is no growth of *Bacillus subtilis* var. globigii colonies on the control plates, use the results for the calculation of the protection factor (see 6.8.3.14).

**6.8.3.12** If there is growth of *Bacillus subtilis* var. globigii colonies on the control plates, clean the equipment and repeat the procedure.

**6.8.3.13** Determine the challenge dose  $N$ , in spores per millilitre, using the equation

$$N = n_2(M_1 - M_2) - (n_2 - n_1)V$$

where

$M_1$  is the mass of the nebulizer plus contents before spraying, in grams;

$M_2$  is the mass of the nebulizer plus contents after spraying, in grams;

$n_1$  is the concentration of the initial suspension before spraying, in spores per millilitre;

$n_2$  is the concentration of the suspension after spraying, in spores per millilitre; and

$V$  is the volume of the initial spore suspension in the nebulizer, in millilitres;

and assuming the density of the spore suspension to be 1,0 g/ml.

#### Example

If the initial mass of the nebulizer was 28,0 g and the final mass 26,8 g, the initial concentration of spores  $4,5 \times 10^8/\text{ml}$  and the final concentration  $4,6 \times 10^8/\text{ml}$  and the initial suspension in the nebulizer 5 ml, the calculation would be as follows:

$$\begin{aligned} N &= 4,6 \times 10^8(28,0 - 26,8) - (4,6 \times 10^8 - 4,5 \times 10^8) \times 5 \\ &= 5,5 \times 10^8 - 0,5 \times 10^8 \\ &= 5,0 \times 10^8 \end{aligned}$$

**6.8.3.14** Calculate a value for the protection factor,  $PF$ , separately for each culture plate using one of the following equations:

a)  $PF = (N.s)/(10n)$ ; or

b)  $PF = (N.s)/(10^4n)$

where

$N$  is the challenge dose;

$s$  is the sampling rate of one slit air sampler (in  $\text{m}^3/\text{min}$ , equation (a), or  $\text{l}/\text{min}$ , equation (b)); and

$n$  is the number of colonies of *Bacillus subtilis* var. globigii on the culture plate.

**Example**

If the challenge dose was calculated as  $4,3 \times 10^8$  spores, the sampling rate was 30 l/min and the number of colonies counted was three, then the protection factor calculated using equation (b) would be as follows:

$$PF = (4,3 \times 10^8 \times 30) / (10^4 \times 3)$$

$$= 4,3 \times 10^5$$

NOTE – When calculating the protection factor using equation (a) or (b) above: if there was only one colony on the sample plate in the example above, the protection factor would be  $1,3 \times 10^6$ . If there were no colonies on the sample plate this would indicate that the protection factor was higher than this, and in the above example the protection factor would be recorded as  $PF > 1,3 \times 10^6$ .

**6.8.3.15** Carry out five replicate protection tests. Ensure that no individual value of the protection factor is less than  $1,0 \times 10^5$  (see 4.2.4.3.2).

**6.8.4 Potassium iodide method****6.8.4.1 Materials and apparatus**

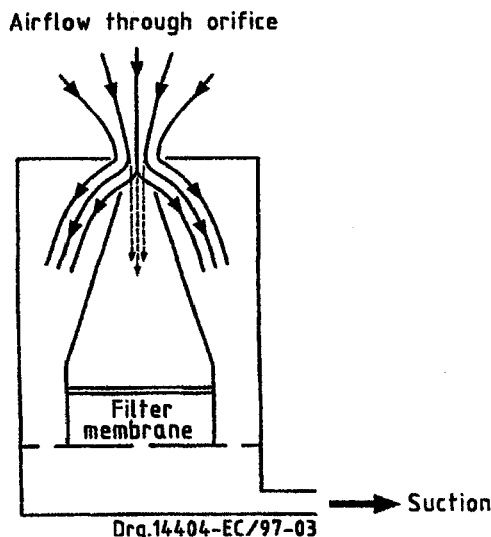
**6.8.4.1.1 Potassium iodide**, 15 g/l solution either in absolute ethanol or industrial methylated spirits with a water content of not more than 5 % (by volume).

**6.8.4.1.2 Palladium chloride**, 1,0 g/l solution in 0,1 mol/l hydrochloric acid.

**6.8.4.1.3 Aerosol generator assembly** that comprises a 3,8 cm diameter spinning disc capable of rotating at 28 000 r/min  $\pm$  500 r/min, and a nozzle with a fine hole to deliver the potassium iodide solution (see 6.8.4.1.1) to the spinning disc, the gap between the end of the nozzle and the spinning disc being set to 0,1 mm; also a laboratory stand to hold the aerosol generator above the work surface when necessary.

**6.8.4.1.4 Air samplers**, that work on a centripetal principle with a volume flow rate of air of 100 l/min through the front orifice, and a cone that entrains some 3 % of this air, collecting approximately 100 % of any potassium iodide particles that enter the sampler; airflow through the samplers being provided by a centrifugal fan coupled to the air samplers by a fixed tube.

NOTE – The particles being heavier than air, follow a straight path through the cone and are deposited on a filter membrane located at the base of the cone, while air is deflected to the outside of the cone (see figure 7).



**Figure 7 — Airflow patterns within the centripetal air samplers**

**6.8.4.1.5 Cylinder** of length and diameter approximately 1 m and 60 mm to 65 mm, respectively, that has a smooth surface and is closed at both ends.

**6.8.4.1.6 Petri dishes**, of diameter 55 mm.

**6.8.4.1.7 Filter membranes**, of diameter 25 mm and with a pore size of 3  $\mu\text{m}$ .

#### **6.8.4.2 Procedure**

**6.8.4.2.1** Set out two Petri dishes (see 6.8.4.1.6) away from the cabinet being tested, one dish half filled with palladium chloride solution (see 6.8.4.1.2) and the other one half filled with distilled water. Replace the lids on each. Set out two filter papers for drying the filter membranes (see 6.8.4.1.7).

**6.8.4.2.2** Introduce the cylinder (see 6.8.4.1.5) through the work-access aperture of the cabinet to mimic the turbulence produced by an operator's arm in front of the aperture. Centre the cylinder between the side walls of the cabinet work space and normal to the plane of the aperture, extending from the back of the work space to protrude at least 250 mm into the room from the plane of the aperture. Raise the lower surface of the cylinder to between 65 mm and 75 mm from the cabinet floor.

**6.8.4.2.3** Place the aerosol generator (see 6.8.4.1.3) inside the work space, with the leading edge of the disc 100 mm behind the plane of the front aperture.

**6.8.4.2.4** For class I cabinets (see figure 6(a)), place the aerosol generator below the cylinder.

**6.8.4.2.5** For class II cabinets (see figure 6(b)), ensure that the disc is level with the upper edge of the aperture.

**6.8.4.2.6** Position two air samplers (see 6.8.4.1.4) outside the cabinet in front of the work-access aperture, with their inlets 150 mm to 160 mm in front of the plane of the aperture. Ensure that the inlets are level with the top of the cylinder, one to the right and one to the left, and each 150 mm from the axis of the cylinder.

**6.8.4.2.7** Load each air sampler carefully with a filter membrane using a pair of fine-pointed forceps kept clean and dry solely for this purpose. Adjust the air pressure to 20 cm water gauge (this is consistent with an air sampling rate of 100 l/min), using a U-tube water manometer one limb of which is attached to a pressure tapping on the rear of the sampler.

**6.8.4.2.8** Measure 20 ml of potassium iodide (see 6.8.4.1.1) into the aerosol generator reservoir with the fluid release valve closed.

**6.8.4.2.9** Switch on the cabinet and allow it to run until normal operating conditions are reached. Apply suction to the air samplers and start the spinning disc. Wait 15 s and then open the release valve to allow the potassium iodide to feed on to the centre of the disc.

**6.8.4.2.10** Turn off the air samplers after the generation of aerosol has stopped. Wait until the suction pump stops completely, and then remove the filter membrane from one sampler using a second pair of fine-pointed forceps designated for this purpose.

**6.8.4.2.11** Gently float the filter membrane in the palladium chloride solution contained in the Petri dish, with the surface that has been exposed to airflow facing upwards. Within 30 s to 45 s, the membrane will become saturated with palladium chloride and any potassium iodide particles will become visible as brown spots.

**6.8.4.2.12** Remove the membrane with a third pair of fine-pointed forceps designated for this purpose, immerse the membrane in distilled water for 3 s to 4 s and then place it on a clean filter paper to dry. Repeat this procedure with the filter membranes from the other air samplers. Replace the lids of the Petri dishes.

NOTE – The solution of potassium iodide used for the tests is flammable and corrosive to untreated steel; consequently

the cabinet under test should be wiped clean with a wet cloth and scrupulous care should be taken with the spinning disc equipment.

**6.8.4.2.13** Examine each filter with either a low-power binocular microscope or a 15 × magnifier and count the number of brown spots on the filter membrane.

#### **6.8.4.2.14 Calculation of the protection factor**

Calculate the number of potassium iodide particles released,  $N$ , using the following equation:

$$N = (3,1 \times 10^7 \times M)$$

where

$3,1 \times 10^7$  is a constant derived from the droplet size, the sampling flow rate and the speed of rotation of the disc; and

$M$  is the volume of potassium iodide solution dispersed by the aerosol generator, in millilitres.

Then calculate a value for the protection factor,  $PF$  (separately for each filter membrane), using the following equation:

$$PF = NV/10^4n$$

where

$V$  is the sampling flow rate in cubic metres per minute; and

$n$  is the number of spots on the filter membranes.

#### **NOTES**

1 In this case,  $M$  is 20 ml (see 6.8.4.2.8) and  $V$  is 100 l/min (see 6.8.4.2.7).

2 Using the above equations and the values of  $M$  and  $V$  given in note 1, a protection factor of  $1,0 \times 10^5$  would correspond to 62 spots on the filter membrane.

3 When calculating the protection factor, if there was only one spot on the filter membrane, the protection factor would be  $2,6 \times 10^6$ . If there were no spots on the filter membrane this would indicate that the protection factor was higher than this, and in the above example the protection factor would be recorded as  $PF > 2,6 \times 10^6$ .

#### **6.8.5 Background tests**

Place two air samplers loaded with filter membranes in front of the safety cabinet, 150 mm to either side of the aperture centre line and 100 mm from the plane of the aperture. Turn on the sampler suction fan and run it for 10 min without any generation of potassium iodide droplets by the aerosol generator.

Remove the filter membranes and develop and examine them in accordance with 6.8.4.2.12 and 6.8.4.2.13.

#### **NOTES**

1 On completion of the background tests in laboratories where no previous tests have taken place within 24 h, the developed membranes should not show any brown spots.

2 In laboratories where protection factor tests have recently taken place (or where protection factor tests have resulted in a considerable leakage of aerosol challenge) it is particularly advisable to perform background tests before further tests on the cabinets. A count of more than five spots on one of the two filter membranes following a 10 min test should be regarded as unsatisfactory and further cabinet tests should be postponed until the background is no longer contaminated.

## **6.9 External contamination test**

### **6.9.1 Principle**

The integrity of the air barrier at the work-access aperture is indicated by the measurement of the inward penetration of bacterial spores which are sprayed into the opening with the cabinet operating normally.

### **6.9.2 Apparatus and materials**

As in 6.8.2.1 (the slit air samplers are not required).

### **6.9.3 Procedure**

**6.9.3.1** Place the cylinder in the cabinet as described in 6.8.3.3.

**6.9.3.2** Distribute at least 12 culture plates (see 6.8.2.1.2) evenly over the work floor of the cabinet.

**6.9.3.3** Load a measured volume of spore suspension into the nebulizer. Position the nebulizer outside the cabinet, with the nebulizer's delivery opening 100 mm in front of the centre of the top edge of the work-access aperture. Ensure that the spray axis is parallel to the work surface and directed into the cabinet.

**6.9.3.4** Ensure that the cabinet is operating normally. Uncover the culture plates 1 min before spraying begins. Run the nebulizer for a period of at least 4 min to ensure the dispersal of a challenge dose of at least  $3 \times 10^6$  spores. Switch off the nebulizer and leave the culture plates uncovered for a further 5 min.

**6.9.3.5** In any test, the number of colonies of the test organism counted after incubation at  $36 \text{ }^{\circ}\text{C} \pm 1 \text{ }^{\circ}\text{C}$  for a period of 24 h to 48 h shall not exceed five. Carry out the test five times, using a new set of culture plates for each test.

**6.9.3.6** Carry out a control test with the cabinet motor blower(s) switched off. Ensure that at least 300 colonies are recovered from these plates during the control test. If less than 300 colonies are recovered, repeat the test.

## **6.10 Cross-contamination test**

### **6.10.1 Principle**

Bacterial spores are sprayed across the work space and contamination of the opposite two-thirds of the cabinet is monitored.

### **6.10.2 Apparatus and materials**

As in 6.8.2.1 (the slit air samplers are not required).

### **6.10.3 Procedure**

**6.10.3.1** Place the cylinder in the cabinet as described in 6.8.3.3.

**6.10.3.2** Distribute at least 12 culture plates (see 6.8.2.1.2) evenly over the right two-thirds of the work surface of the cabinet and at least 350 mm from the left side.

**6.10.3.3** Load a measured volume of spore suspension into the nebulizer. Place the nebulizer with its spray axis 100 mm above the work surface and 50 mm from the left side of the work surface. Ensure that the spray axis is parallel to the work surface and directed towards the opposite wall.

**6.10.3.4** Ensure that the cabinet is operating normally. Uncover the culture plates 1 min before spraying begins and cover them again 5 min after spraying stops. Run the nebulizer for a period of at least 4 min to ensure the dispersal of a challenge dose of at least  $1 \times 10^5$  spores.

**6.10.3.5** Count the number of colonies of the test organism after incubation at  $36\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$  for a period of  $48\text{ h} \pm 2\text{ h}$ . Carry out the test three times.

**6.10.3.6** Carry out the test three more times using reversed positions (i.e. placing the culture plates on the left and the nebulizer on the right).

**6.10.3.7** Carry out a control test with the cabinet motor blower(s) switched off. Ensure that 300 colonies are recovered from these plates during the control test. If less than 300 colonies are recovered, repeat the test.

## **6.11 Test for resistance to corrosion**

### **6.11.1 Test solution**

#### **6.11.1.1 Preparation of the sodium chloride solution**

Dissolve a sufficient mass of sodium chloride in distilled or deionized water of conductivity not higher than  $20\text{ }\mu\text{S/cm}$  at  $25\text{ }^{\circ}\text{C} \pm 2\text{ }^{\circ}\text{C}$  to produce a concentration of the sprayed solution collected of  $50\text{ g/l} \pm 5\text{ g/l}$ . The specific gravity range for a  $50\text{ g/l} \pm 5\text{ g/l}$  solution is 1,025 5 to 1,040 0 at  $25\text{ }^{\circ}\text{C}$ .

Ensure that the sodium chloride contains less than 0,001 % (by mass) of copper and less than 0,001 % (by mass) of nickel as determined by atomic absorption spectrophotometry or another analytical method of similar sensitivity. Ensure that the sodium chloride does not contain more than 0,1 % (by mass) of sodium iodide or more than 0,5 % (by mass) of total impurities calculated for dry salt.

If the pH of the prepared solution, measured at  $25\text{ }^{\circ}\text{C} \pm 2\text{ }^{\circ}\text{C}$ , is outside the range 6,0 to 7,0, investigate for the presence of undesirable impurities in the sodium chloride or in the water (or in both).

#### **6.11.1.2 pH adjustment**

So adjust the pH of the solution that the pH of the sprayed solution collected within the spray cabinet (see 6.11.2.1) is between 6,5 and 7,2. Check the pH using electrometric measurement at  $25\text{ }^{\circ}\text{C} \pm 2\text{ }^{\circ}\text{C}$ , or, in routine checks, with a short-range pH paper which can be read in increments of 0,3 pH units or less. Make any necessary correction by adding hydrochloric acid or sodium hydroxide solution of analytical grade.

Changes in pH can result from the loss of carbon dioxide from the solution when it is sprayed. Such changes can be avoided by reducing the carbon dioxide content of the solution by, for example, heating it to a temperature above  $35\text{ }^{\circ}\text{C}$  before it is placed in the apparatus, or by making the solution from freshly boiled water.

#### **6.11.1.3 Filtration**

If necessary, filter the solution before placing it in the reservoir of the apparatus (see 6.11.2.3), to remove any solid matter which might block the apertures of the spraying device.

### **6.11.2 Apparatus**

Ensure that all components in contact with the spray or the test solution are made of, or lined with, materials resistant to corrosion brought about by the sprayed solution and which, in turn, do not influence the corrosiveness of the sprayed test solutions. The apparatus shall include the components given in 6.11.2.1 to 6.11.2.4.

#### **6.11.2.1 Spray cabinet**

The spray cabinet shall be of a capacity not less than  $0,2\text{ m}^3$  and preferably not less than  $0,4\text{ m}^3$ , since, with smaller volumes, difficulties can be experienced in ensuring the even distribution of spray. For large-capacity cabinets, it is necessary to ensure that the conditions of homogeneity and distribution

of the spray are met (see 6.11.6). The upper parts of the cabinet shall be so designed that drops of sprayed solution formed on its surface do not fall on the specimens being tested.

The size and shape of the cabinet shall be such that the collection rate of solution in the cabinet is within the limits specified in 6.11.6.3.

#### 6.11.2.2 Heater and temperature control

An appropriate system that maintains the spray cabinet and its contents at the specified temperature (see 6.11.6.1). The temperature shall be measured at least 100 mm away from the walls.

#### 6.11.2.3 Spraying device

The device for spraying the sodium chloride solution comprises a supply of clean air at a controlled pressure and humidity, a reservoir to contain the solution to be sprayed, and one or more atomizers.

The compressed air supplied to the atomizers is passed through a filter to remove all traces of oil or solid matter and is at an absolute pressure of 70 kPa<sup>1)</sup> to 170 kPa.

NOTE – Atomizing nozzles might have a "critical pressure" at which an abnormal increase in the corrosiveness of the salt spray occurs. If the "critical pressure" of a nozzle has not been established with certainty, control fluctuations in the air pressure within  $\pm 0,7$  kN/m<sup>2</sup>, by installing a suitable pressure regulator valve to minimize the possibility of the nozzle being operated at its "critical pressure".

In order to prevent the evaporation of water from the sprayed droplets, the air is humidified before entering the atomizer, by passing through a saturation tower that contains hot water at a temperature several degrees Celsius higher than that of the cabinet. The appropriate temperature depends on the pressure used and on the type of atomizer nozzle and these shall be so adjusted that the rate of collection of spray in the cabinet and the concentration of the collected spray are kept within the specified limits (see 6.11.6). The level of the water is automatically maintained to ensure adequate humidification.

The atomizers are made of inert material, for example glass or plastics materials. Baffles may be used to prevent direct impact of spray on the test specimens and the use of adjustable baffles is helpful in obtaining uniform distribution of the spray within the cabinet. The level of the sodium chloride solution in the reservoir is automatically maintained to ensure uniform spray delivery throughout the test.

#### 6.11.2.4 Collecting devices

At least two suitable collecting devices, which consist of funnels made of glass or other chemically inert material and with the stems inserted into graduated cylinders or other similar containers, shall be used. Funnels of diameter 100 mm have a collecting area of approximately 80 cm<sup>2</sup>. The collecting devices are placed in the zone of the cabinet where the test specimens are placed, one close to an inlet of spray and one remote from an inlet. They are so placed that only spray, and not drops of liquid that fall from specimens or from parts of the cabinet, is collected.

#### 6.11.2.5 Re-use

Ensure that if the equipment has been used for a spray test or for any other purpose and a solution different from the specified sodium chloride solution was used, it is thoroughly clean before use.

Operate the equipment for at least 24 h and measure the pH of the collected solution to ensure that it is correct throughout the entire spraying period, before any specimens are placed in the chamber.

#### 6.11.3 Method of evaluation

To check the reproducibility of the test results for one piece of apparatus or for similar items of apparatus in different laboratories, it is necessary to verify the apparatus at regular intervals, as described in 6.11.3.1 to 6.11.3.4.

##### 6.11.3.1 Reference specimens

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1.)  $1 \text{ kN/m}^2 = 0,01 \text{ atm}$ .

To verify the apparatus, use four reference specimens of thickness  $1 \text{ mm} \pm 0,2 \text{ mm}$ , width 50 mm and length 80 mm, of CR4 grade steel that complies with SABS ISO 3574:1986, *Cold-reduced carbon steel sheet of commercial and drawing qualities*, as published by Government Notice No. 399 of 1 April 1999, with a practically faultless surface<sup>2)</sup>, and a matt finish (arithmetical mean deviation of the profile  $R_a = 1,3 \text{ } \mu\text{m} \pm 0,4 \text{ } \mu\text{m}$ ). Cut these reference specimens from cold-rolled plate or strip.

Carefully clean the reference specimens immediately before testing. Besides the directions given in 6.11.4.2 and 6.11.4.3, ensure that the cleaning eliminates all traces of dirt, oil and other foreign matter that could influence the test results.

Use one of the following methods:

- a) clean the reference specimens by vapour degreasing using a chlorinated hydrocarbon. Use three successive treatments of 1 min each, with an interval of at least 1 min between successive treatments; or
- b) thoroughly clean the reference specimens with an appropriate organic solvent (hydrocarbon, that has a boiling point between  $60 \text{ } ^\circ\text{C}$  and  $120 \text{ } ^\circ\text{C}$ ) using a clean soft brush or an ultrasonic cleaning device. Carry out the cleaning in a vessel filled with solvent. After cleaning, rinse the reference specimens with fresh solvent, then dry them; or
- c) other cleaning methods may be used after agreement between the interested parties, provided that the results will be comparable.

Determine the mass of the reference specimens to an accuracy within 1 mg. Protect one face of the reference specimens with a removable coating, for example, an adhesive plastic film.

#### 6.11.3.2 Arrangement of the reference specimens

Position the four reference specimens in four different quadrants in the spray cabinet, with the unprotected faces upwards, and at an angle of  $20^\circ \pm 5^\circ$  from the vertical.

Use supports made of, or coated with, inert materials such as plastics. Ensure that the upper edges of the reference specimens are level with the top of the salt spray collector. The test duration is 96 h.

#### 6.11.3.3 Determination of mass loss

At the end of the test, remove the protective coating. Remove the corrosion products by immersion in a cleaning solution of hydrochloric acid ( $\rho_{20} = 1,18 \text{ g/ml}$ ), of recognized analytical grade at 50 % (by volume), in water, inhibited by 3,5 g of hexamethylene tetramine per litre.

After stripping, thoroughly clean the reference specimens with water at ambient temperature, then with acetone, followed by drying.

Determine the mass of the reference specimens to an accuracy within the nearest 1 mg and calculate the mass loss of the exposed surfaces in grams per square metre.

#### 6.11.3.4 Checking of apparatus operation

The operation of the test apparatus is deemed to be satisfactory if the mass loss of each reference specimen is  $140 \text{ g/m}^2 \pm 40 \text{ g/m}^2$ .

#### 6.11.4 Test specimens

**6.11.4.1** Two rectangular test specimens of nominal dimensions  $100 \text{ mm} \times 15 \text{ mm}$  of the metal used to manufacture the cabinet.

**6.11.4.2** Thoroughly clean the test specimens before testing (see 6.11.3.1). The cleaning method used will depend on the nature of the material, its surface and the contaminants, but abrasives or solvents

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2) "Practically faultless" means free from pores, marks, scratches, and any light coloration.



which might attack the surface of the specimens may not be used.

Ensure that specimens are not recontaminated after cleaning by careless handling.

Specimens intentionally coated with protective organic films should not be cleaned before the test.

**6.11.4.3** If the test specimens are to be cut from a larger coated article, ensure that the coating is not damaged in the area adjacent to the cut. Protect the cut edges by coating them with a material that will be stable under the conditions of the test, such as paint, wax or adhesive tape.

### **6.11.5 Arrangement of the test specimens**

**6.11.5.1** So place the test specimens in the spray cabinet that they are not in the direct line of travel of spray from the atomizer.

**6.11.5.2** The angle at which the surface of a test specimen is exposed in the spray cabinet is very important. Support the specimen facing upwards in the spray cabinet at an angle as close as possible to 20° to the vertical but within the limits 15° to 30°. In the case of irregular surfaces, for example, entire components, adhere to these limits as closely as possible.

**6.11.5.3** So arrange the test specimens that they do not come into contact with the cabinet and that surfaces to be tested are exposed to free circulation of spray. The specimens may be placed at different levels within the cabinet, as long as the solution does not drip from specimens or their supports at one level onto other specimens placed below. However, for a new examination or for tests with a total duration exceeding 96 h, location permutation of specimens is permitted.

**6.11.5.4** Ensure that the supports for the test specimens are made of inert non-metallic material such as glass, plastics or suitably coated wood. If it is necessary to suspend specimens, use synthetic fibre, cotton thread or other inert insulating material.

### **6.11.6 Operating conditions**

**6.11.6.1** Maintain the temperature inside the spray cabinet at 35 °C ± 2 °C with the minimum possible fluctuation in temperature throughout the duration of the test.

**6.11.6.2** Start the test after it has been confirmed that the collection rate (see 6.11.6.3) and conditions (see 6.11.6.1) are within the specified ranges, and the cabinet is filled with test specimens as planned.

**6.11.6.3** Ensure that the solution collected in each of the collecting devices (6.11.2.4) has a sodium chloride concentration of 50 g/l ± 5 g/l and a pH value in the range 6,5 to 7,2.

Ensure that the average rate of collection of solution in each device, measured over a period of at least 24 h of continuous spraying is 1 ml/h to 2 ml/h for a horizontal collection area of 80 cm<sup>2</sup>.

**6.11.6.4** Do not re-use test solution which has been sprayed.

**6.11.6.5** During the test, prevent any increase or decrease of cabinet pressure.

### **6.11.7 Duration of test**

**6.11.7.1** The duration of the test shall be at least 480 h.

**6.11.7.2** Do not interrupt the spraying during the prescribed test period. Do not open the cabinet except for brief visual inspections of the test specimens in position and for replenishing the salt solution in the reservoir, if such replenishment cannot be carried out from outside the cabinet.

**6.11.7.3** A periodic visual examination of specimens under test for a predetermined period is allowed, but do not disturb the surfaces under test and only open the cabinet for the minimum period necessary

to observe and record any visible changes of the material under test.

#### **6.11.8 Treatment of specimens after test**

At the end of the test period, remove the test specimens from the cabinet and allow them to dry for 0,5 h to 1 h before rinsing, in order to reduce the risk of removing corrosion products. Before they are examined, carefully remove the residues of spray solution from the surfaces of the test specimens. A suitable method is to rinse or dip the test specimens gently in clean running water, at a temperature not exceeding 40 °C and then to dry them immediately in a stream of air, at a pressure not exceeding 200 kPa and at a distance of approximately 300 mm.

#### **6.11.9 Evaluation of results**

The material shall be deemed to be corrosion resistant if corrosion of the base metal is not visible to the unaided eye.

#### **6.11.10 Report**

**6.11.10.1** The report shall indicate the outcome of the test according to the criteria for the evaluation of results. Report the result obtained for each specimen tested and, when appropriate, the average result for a group of replicate test specimens. The report could be accompanied by photographic records of the tested specimens.

**6.11.10.2** The following information shall be included in the report:

- a) a reference to this compulsory specification;
- b) the type and purity of sodium chloride and water used;
- c) a description of the material or product tested;
- d) the dimensions and shape of the test specimen and the nature and area of the surface tested;
- e) the preparation of the test specimen, including any cleaning treatment applied and any protection given to edges or other special areas;
- f) known characteristics of any coating with an indication of the surface area;
- g) the number of test specimens subjected to the test and a reference to each material or product represented;
- h) the method used to clean test specimens after the test, with, where appropriate, an indication of the loss in mass resulting from the cleaning operation;
- i) the angle at which the tested surfaces were inclined;
- j) the frequency and number of specimen location permutations, if any;
- k) the duration of the test and the results of any intermediate inspections;
- l) the test temperature;
- m) the volume of collected solution;
- n) the pH of the test solution and the collected solution;
- o) the density of the collected solution;
- p) any abnormality or incident that occurred during the entire test procedure; and

q) intervals of inspection.

## 6.12 Prefilter tests

Use the relevant test methods given in SABS 1424, *Filters for use in air-conditioning and general ventilation*, as published by Government Notice No. 1851 of 1 December 1995, to determine the initial arrestance.

Use a 600 mm × 600 mm sample of the prefilter in its unpleated state.

## 6.13 Chemical resistance

### 6.13.1 Reagents

Use the following reagents for the testing of chemical resistance:

- hydrochloric acid, 40 g/ℓ<sup>3)</sup>
- sodium hydroxide, 40 g/ℓ
- quaternary ammonium compound, 10 g/ℓ
- formaldehyde, 50 g/ℓ
- sodium hypochlorite, 5 g/ℓ
- iodophor, 20 g/ℓ
- phenol, 50 g/ℓ
- ethanol, 70 % (by volume)

### 6.13.2 Procedure

**6.13.2.1** Apply approximately 0,5 ml of each reagent to the surface that has to be tested. Cover each reagent with a watch glass in the centre of the puddle with the concave side down and leave the reagent on the surface for 4 h, ensuring that the test surface is wet throughout the entire period.

**6.13.2.2** After the 4 h period, scrub the surface with a stiff brush and hot water at a temperature of 70 °C.

**6.13.2.3** Dry the surface and remove any surface stains by washing with alcohol before examination.

### 6.13.3 Inspection

Inspect the test surfaces for any visible effect other than a slight change of gloss or discoloration when compared with an untreated area.

## 6.14 Stability test

### 6.14.1 Principle

The stability test is performed to demonstrate the resistance of a microbiological safety cabinet to overturning under an applied force.

### 6.14.2 Apparatus

**6.14.2.1** Compression force gauge, calibrated in kilograms, accurate to within ± 5 % full scale, or

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3) For example: Use 125 ml of concentrated hydrochloric acid (at a concentration of 32 % (by mass)) and dilute to 1 ℓ.

**6.14.2.2 Extension spring balance**, calibrated in kilograms, accurate to within  $\pm 5\%$  full scale.

### 6.14.3 Procedure

**6.14.3.1** Block the cabinet at the front bottom edge to prevent lateral movement. Apply force, as appropriate, at the centre of the rear top edge (see figure 8) to subject the cabinet to a torque of 700 Nm.

**6.14.3.2** Measure the cabinet lift at the rear bottom edge.

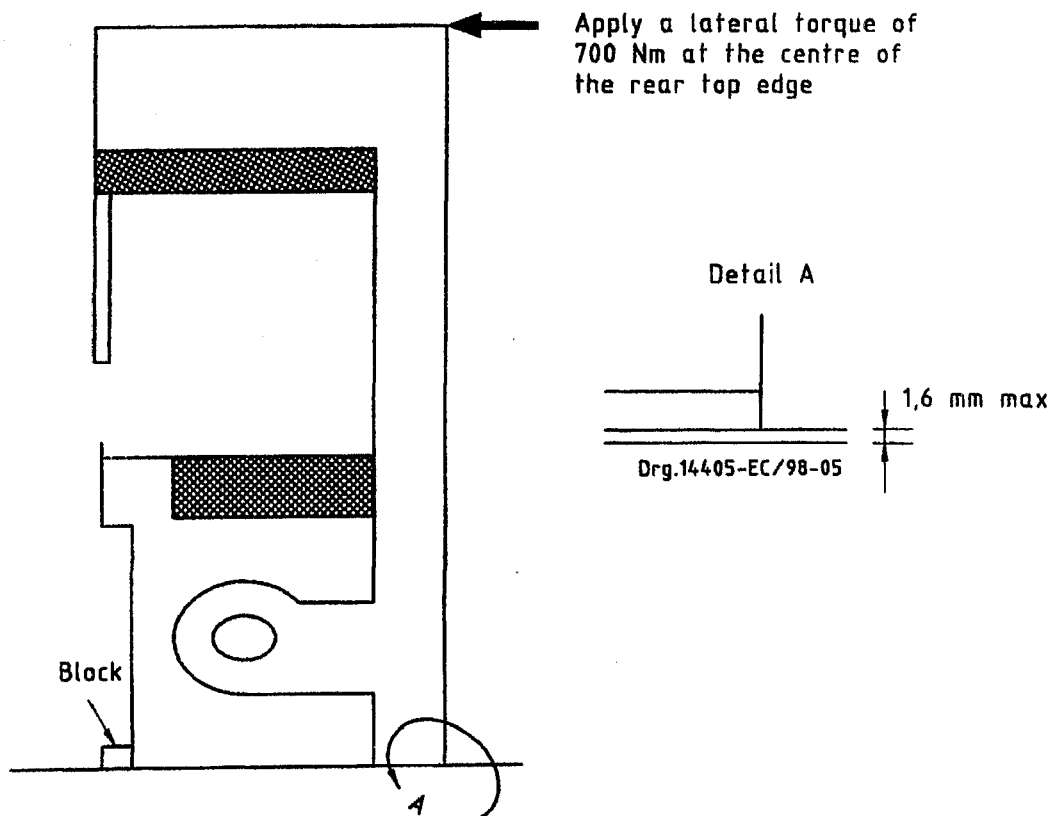


Figure 8 — Resistance to overturning

## 7 Blower performance test

### 7.1 Principle

The airflow velocity is increased to determine whether the motor blower has spare capacity.

### 7.2 Apparatus

**7.2.1 Vane anemometer** or **thermo-anemometer** (or both) as appropriate, accurate to within 2 %.

**7.2.2 Timer.**

### 7.3 Procedure

#### 7.3.1 Class I

**7.3.1.1** With clean HEPA-filters fitted, set the blower speed control so that when the inward airflow

velocity is measured in accordance with 6.6.3.1, the average airflow velocity shall be that specified in 3.6.2.1.

**7.3.1.2** After 2 h, measure the average inward airflow velocity in accordance with 6.6.3.1.

### **7.3.2 Class II**

**7.3.2.1** With clean HEPA-filters fitted, set the blower speed control so that when the downward airflow velocity in the work space is measured in accordance with 6.6.3.2.1, the average downward airflow velocity shall be that specified in 3.6.2.2.

**7.3.2.2** After 2 h, measure the average downward airflow velocity in accordance with 6.6.3.2.1.

**7.3.2.3** When a class II cabinet is fitted with a separate motor blower for the exhaust system, set the blower speed control so that when the inward airflow velocity through the work-access aperture is measured in accordance with 6.6.3.2.2, the average inward airflow velocity shall be that specified in 3.6.2.2.

**7.3.2.4** After 2 h, measure the average inward airflow velocity in accordance with 6.6.3.2.2.

## **8 Documentation, information and marking**

### **8.1 Documentation and information**

The following documentation and information shall be provided by the manufacturer:

- a) installation instructions, including recommendations as to the siting (location) of the cabinet in the laboratory;
- b) operating instructions;
- c) maintenance instructions that shall include at least the following:
  - 1) instructions for the maintenance and replacement of filters, including a statement of the need for appropriate decontamination of the cabinet;
  - 2) reference to the need to clean and disinfect surfaces in the work zone at regular intervals;
  - 3) a warning regarding cleaning agents and other materials known to be incompatible with sealing materials and finishes;
  - 4) a reference to the size of the HEPA-filters; and
  - 5) a reference to appropriate performance checks; the frequency of such checks and the acceptance/rejection criteria for such checks, and the test methods.

### **8.2 Marking**

For each class of cabinet, the appropriate of the following information shall be prominently, legibly and indelibly displayed on the front of each cabinet:

- a) the designation, i.e.
  - 1) Microbiological safety cabinet class I – Protection for personnel and the environment against ordinary microbiological agents, or
  - 2) Microbiological safety cabinet class II – Protection for personnel, the environment and the product against ordinary microbiological agents, or
  - 3) Microbiological safety cabinet class III – Protection for personnel, the environment and the product against special and extremely hazardous microbiological agents;

- b) the manufacturer's name and address;
- c) the total volume of the cabinet;
- d) the words **"WARNING: Do not use flammable or explosive and highly volatile liquids in this cabinet"**;
- e) the serial number of the cabinet;
- f) the year of manufacture; and
- g) the electrical voltage (volts), frequency (hertz) and current drawn (amps).